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# Annals of the Missouri Botanical Garden

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No. 1

# A REVISION OF THE GENUS PRIVA<sup>1</sup>

#### CLARENCE EMMEREN KOBUSKI

Rufus J. Lackland Research Fellow in the Henry Shaw School of Botany of Washington University

# HISTORY OF THE GENUS

The tropical genus *Priva* was established by Adanson<sup>2</sup> in the 'Familles des Plantes' in 1763. The genus was segregated from *Verbena* on account of the characters of the fruit and was based on a single species, namely, *Verbena lappulacea*, published by Linnaeus<sup>3</sup> in the 'Species Plantarum' in 1753.

Jussieu<sup>4</sup> in 'Observations sur la Famille des Plantes Verbenacees,' published in 1806, accepted Adanson's genus *Priva* and transferred thereto several species, including *P. dentata*, *P. echinata*, *P. hispida*, *P. laevis*, and *P. leptostachya*. In the following year Persoon<sup>5</sup> gave a brief synopsis of the genus, recognizing the five species listed by Jussieu, but made no further additions to the group.

Humboldt, Bonpland and Kunth, in 1817, added a new species, P. aspera, from material collected in Mexico. During the following three decades five additional species were described by different authors. They were P. crenata Schrad., P. Forskalii

- <sup>1</sup> An investigation carried out at the Missouri Botanical Garden in the Graduate Laboratory of the Henry Shaw School of Botany of Washington University and submitted as a thesis in partial fulfillment of the requirements for the degree of master of science in the Henry Shaw School of Botany of Washington University.
  - <sup>2</sup> Adanson, Fam Pl. 2: 505. 1763.
  - \*Linnaeus, Sp. Pl. ed. 1. 28. 1753.
  - <sup>4</sup> Jussieu, Ann. Mus. Paris 7: 70. 1806.
  - <sup>5</sup> Persoon, Syn. Pl. 2: 139. 1807.
  - HBK, Nov. Gen. & Sp. 2: 278. 1817.
  - <sup>7</sup> Schrader, Linnaea 8, Litteratur Bericht: 24. 1833.
  - Jaubert & Spach, Ill. 5: 59, pl. 455. 1842.

lssued May 8, 1926.

Jaub. & Spach, P. orchioides¹ Walp., P. trachelioides² Mart. & Gal., and P. lamiifolia³ Mart. & Gal. Thus in Walpers⁴ 'Repertorium Botanices Systematicae,' published in 1844–1847, more or less complete descriptions were given for ten recognized species.

The first comprehensive monographic treatment of the genus *Priva* was by Schauer<sup>5</sup> in De Candolle's 'Prodromus' in 1847. In this treatment several previously published species were reduced to synonymy and one new species, *P. bahiensis* DC. from Brazil was described. Since 1847, although several species, namely, *P. tuberosa*<sup>5</sup> and *P. armata*<sup>7</sup> Watson, *P. portoricensis*, and *P. domingensis*<sup>9</sup> Urban have been published, yet no comprehensive study of the group as a whole has been made.

During recent years the numerous botanical expeditions, particularly to the West Indies, Central and South America, have resulted in the accumulation of a comparatively large representation of many of the smaller genera. In organizing the material of the small genus *Priva* in the herbarium of the Missouri Botanical Garden, although a relatively large number of specimens was available for study, yet considerable difficulty was met in the identification of species as well as in the determination of the proper specific names to be used. Hence the present investigation was undertaken to determine the limitation and geographical distribution of species and to establish the correct names to be used.

# GENERAL MORPHOLOGY

Roots.—The root system in the genus Priva presents comparatively little variation. Most of the species develop a rather persistent, more or less branched tap-root. P. rhinanthifolia with

- <sup>1</sup> Walpers, Rep. 4: 36. 1844-1848.
- <sup>2</sup> Mart. & Gal. Bull. Acad. Brux. 11<sup>2</sup>: 324. 1844.
- <sup>1</sup> Ibid. Bull. Acad. Brux. 11<sup>2</sup>: 324. 1844.
- <sup>4</sup> Walpers, Rep. 4: 34. 1844-1848; and 6: 687. 1846-1847.
- <sup>6</sup> Schauer in DC. Prodr. 11: 533. 1847.
- <sup>6</sup> Watson, Proc. Am. Acad. 18: 135. 1882–1883.
- <sup>7</sup> Ibid. Proc. Am. Acad. 25: 160. 1890.
- 8 Urban, Symb. Ant. 4: 534. 1903.
- <sup>9</sup> Ibid. Symb. Ant. 7: 354. 1912.

distinctly tuberous roots is a notable exception, and constitutes therefore an outstanding element in the genus.

Stems.—The stem is typically that of an herbaceous perennial with a woody base. There is considerable variation in size, however, from the low, more or less decumbent stem of P. rhinanthifolia to the stout, erect stem of P. aspera which grows to a height of 18 decimeters. The stem may be simple or branched. In the case of P. armata, the branching is from the base and is usually arcuous-ascending. In all cases the stem is square and often somewhat furrowed or slightly striated.

Leaves.—The variation of the leaf characters within the genus is very great. There is a gradation in length from 1.5 centimeters, as found in *P. portoricensis*, to that of 10 centimeters, as shown by *P. aspera*. The general outline of the leaf, in the majority of species, is more or less of the ovate, subcordate type. *P. rhinan-thifolia* presents the only exception. In this species the outline of the leaf is distinctly oblong. All the species have leaves which are quite regularly dentate and pubescent; in some cases, however, the pubescence is strigose. Both petiolate and sessile leaves occur, with the former type the more common.

Inflorescence.—The inflorescence is a spicate raceme throughout the whole genus, varying greatly, however, in length. In the majority of cases the flowers are pedicellate and solitary. The length and the curvature of the pedicel are important points in the delimitation of the species within the genus.

Calyx.—In antithesis variation in the calyx is not very marked. The persistent calyx, however, presents characters most helpful for specific determination. The calyx may be inconspicuously lobed and thus terminate rather abruptly, or it may be distinctly lobed and the lobes contorted, coarctate, and decidedly beaked. The pubescence, usually present and of straight or uncinate-hispid hairs, plays an important role in classification.

Corolla.—The corolla, in most cases, is deciduous and slightly bilabiate. The tube shows diversity in that it may barely exceed the calyx in length, or it may be two or three times as long. Some instances show the tube and throat to be pubescent but in most cases pubescence is absent.

Stamens.—The stamens are didynamous, inserted at unequal

levels in the corolla-tube and included or nearly so. The lateroanterior pair is more highly developed and inserted a little above the middle of the tube. The two smaller latero-posterior stamens are inserted approximately at the middle of the tube. The posterior stamen is rudimentary or entirely lacking. The anthers are two-celled and the cells are parallel or slightly divergent.

Pistil.—The pistil is typically bicarpellate throughout the whole genus with an ovary which, in the majority of cases, is two-celled. P. mexicana and P. aspera possess a one-celled ovary, due to the abortion of the posterior cell. The style is included and usually glabrous. The stigma is two-lobed, the posterior lobe, which is somewhat reduced, is tooth-like, while the anterior lobe is club-shaped.

Cocci.—The cocci, although varying considerably in shape and size, furnish relatively constant specific characters which aid greatly in specific diagnosis. The dorsal surface may be echinate or ridged. In the latter case the fruit is usually ovate. The commissural surface may be excavated, margined, plane or nearly so. The cocci in most cases are two-celled and two-seeded.

# GEOGRAPHICAL DISTRIBUTION

The genus *Priva* is essentially a tropical genus. The majority of species are found between 30° N. and 35° S. latitude. Of the eleven recognized species, nine occur exclusively in the western hemisphere, while the other two are found in the eastern hemisphere only. Two species are confined to South America. *P. bahiensis* is known only from Brazil, while the range of *P. cuneato-ovata* does not seem to extend north of Argentina and Chili. The species *P. rhinanthifolia*, aspera, and mexicana are indigenous to Mexico and Central America.

The only species found in the United States is the type species, *P. lappulacae*, which is rather cosmopolitan in tropical America.

The African species, *P. leptostachya*, seems to offer the greatest problem in distribution. It is possible, however, that this species as here interpreted may be too inclusive; but the material at hand is inadequate, lacking either foliage, flowers, or fruit to permit of satisfactory or final treatment. A survey of the ma-

terial at hand, however, shows the distribution of this species to extend from the island of Socotra south to, and including, Cape Colony.

The geographical distribution of the genus, as well as the distribution of individual species, is indicated in pl. 1.

# ACKNOWLEDGMENTS

The writer wishes to take this opportunity to express his appreciation and gratitude to the several people who have made the present study possible. Thanks are due Dr. George T. Moore. Director of the Missouri Botanical Garden, for the use of the excellent library and herbarium facilities which this institution Sincere appreciation is due Mr. W. R. Maxon, of the United States National Herbarium, Dr. B. L. Robinson, of the Gray Herbarium, and Mr. D. C. Davies, Director of the Field Museum, who so willingly loaned material from the abovementioned herbaria, and to Professor J. Paul Goode, of the University of Chicago, for permission to use his Homolosine Equal Area Projection Map No. 101 HC. Especial thanks are due to Dr. J. M. Greenman, Curator of the Herbarium of the Missouri Botanical Garden, under whose guidance this revision has been completed, for the aid and advice which were so willingly given at all times.

#### ABBREVIATIONS

The abbreviations used, to indicate the herbaria in which the specimens cited in the present paper occur, are as follows: C = University of Chicago Herbarium (deposited in the Field Museum); F = Field Museum of Natural History Herbarium; G = Gray Herbarium of Harvard University; M = Missouri Botanical Garden Herbarium; US = United States National Herbarium.

## TAXONOMY

Priva Adans. Fam. Pl. 2: 505. 1763; Persoon, Syn. Pl. 2: 139. 1807; Schauer in DC. Prodr. 11: 532. 1847; Bocquillon, Rev. Verb. 115. 1861–1863, excl. Dipyrena; Bentham & Hooker, Gen. Pl. 2: 1145. 1873–1874; Briquet in Engl. & Prantl, Nat.

Pflanzenfam. IV. Abt. 3a, 155. 1897; Lam, Verb. Malayan Arch. 23. 1919.

Blairia Houst. ex Linn. Gen., ed. 1, 334. 1737

Phryma Forsk. Fl. Aegypt. Arab. 115. 1775, not L.

Streptium Roxb. Pl. Corom. 2: 25. t. 146. 1798.

Tortula Roxb. ex Willd. Sp. Pl. 3:359. 1800.

Pitraea Turcz, in Bull. Soc. Nat. Moscow 352: 328. 1862.

Phelloderma Miers in Trans. Linn. Soc. London 27: 100. 1870.

Herbaceous caulescent perennials, glabrous or pubescent. Leaves opposite, sessile or petioled, membranaceous, dentate. Inflorescence spicate, terminal or axillary. Flowers small, solitary, and axillary. Bracts small, lanceolate to ovate. Calyx tubular in anthesis, 5-ribbed, terminating in 5 short teeth, persistent, enlarging with and investing the fruit, usually contracted at the orifice at maturity. Corolla-tube cylindrical; limb spreading, oblique, slightly bilabiate. Stamens 4, didynamous, adnate to about the middle of the corolla-tube, included or nearly so; anther cells parallel or slightly divergent; posterior stamen or staminode much reduced, minute or absent. Ovary 2-celled; ovules 2 or by abortion 1; stigma 2-lobed; posterior lobe tooth-like, anterior lobe club-shaped. Fruit included in the enlarged calyx, separating at maturity into 2-celled (or by abortion 1-celled) cocci. Pericarp hard: dorsal surface echinate, ridged, or smooth: commissural surface excavated, concave, or plane.

Type species: P. lappulacea (L.) Pers. Syn. Pl. 2: 139. 1807.

### KEY TO THE SPECIES

A. Dorsal surface of cocci distinctly echinate.

- D. Leaves ovate, .5-3 cm. long, .3-2 cm. wide; fruit strongly contracted at the base.
- DD. Leaves ovate, 1.5-10 cm. long, 1-6 cm. wide; fruit not strongly contracted at the base.
  - F. Commissural surface of cocci excavated; corolla at least twice the length of the calyx.
    - G. Corolla-tube spirally contorted in bud......4. P. leptostachya

- - HH. Stem and leaves more or less hirsute; Mexican species.
    - I. Leaves sessile, oblong, sharply dentate toward the apex.

9. P. rhinanthifolia

- II. Leaves petioled, ovate, subcordate, uniformly dentate.

  - JJ. Fruiting calyx densely uncinate-hispid, not beaked, reflexed.

# 1. Priva Curtisiae Kobuski, n. sp.1

Herbaceous perennial; stem erect or somewhat decumbent, quadrangular, pubescent, striate, branched, 30-50 cm. high; leaves sessile, ovate to oblong, 0.5-4.5 cm. long, 0.5-2.5 cm. broad, obtuse at the apex, truncate or nearly so at the base, crenate-serrate, scabrous above, paler and somewhat pubescent beneath; inflorescence terminal, spicate, racemes 8-24 cm. long, loosely flowered, pedunculate; bracts ovate to lanceolate, 1-2 mm. long, covered with a fine pubescence; calvx tubular in anthesis, 5-7 mm. long, teeth short, obtuse, fruiting calvx subglobose, 5 mm. in diameter, densely pubescent with uncinate-hispid hairs intermingled with a straight pubescence, basal portion dilated. enclosing the fruit, apical portion somewhat connivent at the orifice; corolla white, 8-10 mm. long, tube spirally twisted, limb slightly bilabiate, lobes rounded; fruit consisting of 2 bilocular cocci, dorsal surface convex, covered with many stout, pubescent spines, commissural surface deeply excavated, margined.

¹ Priva Curtisiae sp. nov., herbaceis perennis; caule erecto vel plus minusve decumbente, ramoso, 30–50 cm. alto, quadrangulare, pubescente, striato; foliis sessilibus, ovatis vel oblongis, 0.5–4.5 cm. longis, 0.5–2.5 cm. latis, obtusis, basi truncatis vel subcordatis, erenato-serratis, supra scabris, subtus pallioribus et hirsuto-pubescentibus; inflorescentiis terminalibus, spicato-racemosis, 8–24 cm. longis, pedunculatis, floribus remotis, pedicellis 1–3 mm. longis; bracteis ovatis vel lanceolatis, 1–2 mm. longis, tenuiter pubescentibus; calyce anthesi tubulosi, 5–7 mm. longo, lobis subobsoletis, calyce fructifero subrotundo, inflato 5 mm. diametro, apice connivente; corolla alba, 8–10 mm. longa, spiraliter contorta, limbo parve bilabiato, lobis rotundatis; capsula late ovata vel subrotunda, matura septicide secedens in coccos duos; coccis bilocularis, dorso convexo grosse multisque pubescentibus spinosis testis; commissura coccorum alte excavata marginata.

Distribution: Kenya Colony, British East Africa.

Specimens examined:

British East Africa: in dry, light soil, Loito Plains, Kenya Colony, 1500-2100 m. alt., 6 June 1923, Curtis 499 (G, TYPE, M, fragment and photograph); hillside in hard, dry soil, open to sun, Loito Plains, Kenya Colony, 1500-2100 m. alt., 28 June 1923, Curtis 597 (G); Loito Plains, Kenya Colony, 1500-2100 m. alt., 3 July 1923, Curtis 642 (G); in wet soil, but after rain usually dry, Mau Range, Kenya Colony, 1500-2100 m. alt., 31 May 1923, Curtis 472 (G).

This species is related to *P. leptostachya* Juss. but differs in having sessile leaves, and evenly echinate and uniformly larger fruit. The species is dedicated to *Mrs. Anita Grosvenor Curtis*.

# 2. Priva portoricensis Urban, Symb. Ant. 4:534. 1903.

A slender, branched perennial 30-40 cm. high; stem woody below, puberulent; leaves petiolate, deltoid to ovate, 0.5-3 cm. long, 0.3-1.5 cm. broad, somewhat crenate-serrate, acute to obtuse at the apex, nearly truncate at the base, pubescent; racemes 25 cm. or less in length; pedicels 1-1.5 mm. long; calyx 5 mm. long, covered with dense minute hairs, lobes triangular; fruiting calyx somewhat rotund; corolla pale blue, tube 6 mm. long, rising well above the calyx, slightly ampliated at the throat, lobes obovate-rotund; fruit cuneate or obcordate, strongly contracted at the base; cocci bearing two rows of spines on the dorsal surface, 2-celled, commissural surface excavated.

Distribution: Porto Rico.

Specimens examined:

Porto Rico: in thickets near Guanica, 2 Feb. 1886, Sintenis 3597, co-type (US, G).

# 3. Priva domingensis Urban, Symb. Ant. 7: 354. 1913.

Stem 10-20 cm. high, branched below, branches glabrous or sparingly pubescent; leaves petiolate, ovate, 5-15 mm. long, 4-8 mm. wide, truncate at the base and somewhat decurrent on the petiole, obtuse to rotund at the apex, often shortly apiculate, crenate, or rarely subentire, finely pubescent; racemes 6 cm. or less in length with a peduncle 1-3 cm. long; flowers few, 2-5,

pedicels 1.5 mm. long; calyx 5 mm. long, covered with sparse short hairs intermixed with dense hooked hairs, lobes short, widely triangular; corolla violet-red, 11.5 mm. long, nearly twice as long as the calyx, somewhat ampliated above, lobes rounded.

Distribution: San Domingo at La Vuelta, near the river Las Lavas, in lime hills.

No specimens seen. Description translated from the original.

4. Priva leptostachya Juss. Ann. Mus. Paris 7: 70. 1806; Pers. Syn. Pl. 2: 139. 1807; Walpers, Rep. 4: 35. 1844; Schauer in DC. Prodr. 11: 532. 1847; Bocquillon, Rev. Verb. 116. 1861–1863; Clark in Hooker, Fl. Brit. Ind. 4: 565. 1885; Pearson in Fl. Capensis 5: 206. 1901; Lam, Verb. Malayan Arch. 24. 1919.

Tortula aspera Roxb. in Willd. Sp. Pl. 3:359. 1801.

- P. dentata Juss. Ann. Mus. Paris 7: 70. 1806; Pers. Syn. Pl.
  2: 139. 1807; Schauer in DC. Prodr. 11: 533. 1847.
- P. abyssinica Jaub. & Spach. Ill. Pl. Orient. 5: 58. t. 453. 1853-57.
- P. Forskalii Jaub. & Spach. Ill. Pl. Orient. 5: 59. t. 455. 1853-57.
  - P. Forskaolafi E. Mey. Comm. Pl. Afr. Austr. 12:75. 1837.
  - P. Meyeri Jaub. & Spach. Ill. Pl. Orient. 5: 57. 1853-57.

An herbaceous perennial 3-9 dm. high, branched; stem 4-sided, striate; leaves petiolate, ovate, 2-11 cm. long, 1-5 cm. wide, coarsely crenate-serrate, rounded or obtuse at the apex, cuneate at the base, strigosely pubescent on both surfaces; racemes terminal or axillary, elongate, 2-3 dm. long; flowers many, distant, shortly pedicellate; calyx in anthesis cylindrical, 3-6 mm. long, in fruit globose, uncinate-hispid; corolla white, bilabiate, twice as long as the calyx, twisted in anthesis; fruit ovate, obcordate, hard, glabrous or sometimes pubescent, composed of two slightly coherent 2-celled cocci, longitudinally ridged with two rows of short spines, commissural surface deeply excavated.

Distribution: in grassy plains and river banks, Island of Socotra to South Africa, India, and East Indies.

Specimens examined:

AFRICA: Island of Socotra, Feb.-March 1880, Balfour 542 (G)

doubtfully referred to this species; vicinity of Kampala, on the trail from Entebbe, Victoria Nyanza to Butiaba, Albert Nyanza, Uganda, 650–1110 m. alt., 21–22 Dec. 1909, Mearns 2402 (US); Ripon Falls, Uganda Protectorate, 18 July 1913, Dümmer 30 (US); Nyasaland, Rhodesia, 1891, Buchanan 887 (US); Mozambique, exact locality and date of collection lacking, Howard 118 (US); River Shire, British Nyasaland Protectorate, coll. of 1863, Kirk (G); Durban, Union of South Africa, March 1894, Kuntze (US, 633155); in fields and mountains near Enon, South Africa, 450 m. alt., Drege a (M); in woods, Somerset, Cape Colony, 750 m. alt., March 1886, Bolus 306 (F); exact locality and date of collection lacking, Burchell 3625 (G); Boschberg, 900 m. alt., without date, MacOwan (F).

East Indies: locality and date not indicated, probably collected by Wallich (M, 119874).

Asia: in the locality of Maisur and Carnatic, British India, without further data, *Thomson* (G).

# 5. Priva bahiensis DC. Prodr. 11: 533. 1847.

Stem quadrangular, narrow, branched, 1.5-3 dm. high, finely pubescent; leaves shortly petiolate, ovate, subcordate, 2.8-4 cm. long, 1-2 cm. wide, coarsely serrate, narrowed at the base into the petiole, strigosely pubescent above, finely pubescent beneath; racemes terminal, 2 dm. long, flowers solitary, in the axils of the bracts, distant; bracts 3-4 mm. long, about twice as long as the pedicels, somewhat linear, pubescent; calyx slender, tubular, 5-ribbed, densely uncinate-hispid, especially between the ribs, 4-5 mm. long, accrescent, becoming broadly inflated, globose, splitting in halves, connivent at the orifice; corolla twice the length of calyx; fruit obcordate, attenuate at base, splitting into 2 bilocular cocci at maturity, dorsal surface convex, possessing two rows of long, slender, curved spines, transversely ridged between the two rows of spines, commissural surface excavated and margined.

Distribution: eastern Brazil.

Specimens examined:

Brazil: along fences and near water, Province of Bahia, Salzmann (M, 118802).

6. Priva lappulacea (L.) Pers. Syn. Pl. 2: 139. 1807; Kuntze, Rev. Gen. Pl. 2: 509. 1891; Rusby, Bull. Torr. Bot. Club 27: 80. 1900; Urban, Symb. Ant. 4: 534. 1903; Britton, Fl. Bermuda, 313. 1918; Britton & Millsp. Bahama Fl. 367. 1920; Urban, Symb. Ant. 8: 597. 1921.

Verbena lappulacea L. Sp. Pl. 28. 1758.

Priva echinata Juss. Ann. Mus. Paris 7: 70. 1806; Kunth, Syn. Pl. Aeq. 2: 61. 1823; Walpers, Rep. 4: 34. 1844–1847; Schauer in DC. Prodr. 11: 534. 1847; Bocquillon, Rev. Verb. 116. 1861–1863; Griseb. Fl. Br. W. Ind. 492. 1864; Griseb. Cat. Pl. Cubensis, 215. 1866; Gray, Syn. Fl. N. Am. 2<sup>1</sup>: 324. 1878; Small, Fl. Southeastern U. S., ed. 2, 1013. 1913.

P. lamiifolia Mart. & Gal. Bull. Acad. Brux. 11<sup>2</sup>: 325. 1844. Stem erect, simple or branched, quadrangular, 2-6 dm. high, pubescent; leaves petiolate, ovate, subcordate, 2.5-11 cm. long, 1-6 cm. wide, coarsely dentate, slightly acuminate at the apex, truncate to subcordate at the base, strigosely pubescent; racemes loosely flowered, 8-18 cm. long; flowers pedicellate; calyx in anthesis tubular, 2-3 mm. long, densely uncinate-hispid, fruiting calyx broadly ovate, coarctate at the apex; corolla slightly surpassing the calyx, blue, pink or white, salverform, oblique, slightly bilabiate, 5-lobed, lobes small, rotund; fruit consisting of 2 bilocular cocci, quadrangular, dorsal surface echinate, scrobiculate between the spines, commissural surface plane or nearly so.

Distribution: cosmopolitan weed of tropical America and the West Indies; in sandy soil, along railroad tracks, open fields, and rocky places.

Specimens examined:

UNITED STATES:

FLORIDA: Key West, Aug. 1877, Garber (US, G); Key West, date lacking, Blodgett (US, G); Key West, 1874, Ed. Palmer 395 (C, US, M); exact locality lacking, coll. of 1842–1849, Rugel (M). MEXICO:

Lower California: San José del Gabo, 15 Sept. 1890, Brandegee (US, G).

Tamaulipas: vicinity of Victoria, 320 m. alt., 1 May-13 June 1907, Ed. Palmer 502 (US, G, and F); near Tampico, 15 m. alt., 1-31 Jan. 1910, Ed. Palmer 8 (G, US, M).

SINALOA: Culiacan, 1891, Ed. Palmer (US); Culiacan, 12 Oct. 1904, Brandegee (US); Culiacan, 27 Aug.—15 Sept. 1891, Ed. Palmer 1458 (G); foothills of Sierra Madre, near Colomas, 21 July 1897, Rose 3240 (US); foothills of Sierra Madre, near Colomas, 16 July 1897, Rose 1722 (US); rancho del Agua Fria, San Ignacio, 430 m. alt., 12 June 1918, Montes 389 (US); dry hills near Mazatlan, 30 March 1910, Rose 13711 (US).

Jalisco: Manzanillo, 1-31 Dec. 1890, Ed. Palmer 1007 (US). Colima: coffee plantation northwest of Colima, 540 m. alt., 28 July 1905, Goldsmith 89 (G).

Guerrero: Iguala, 10-12 Aug. 1905, Rose 9419 (US); vicinity of Acapulco, Oct. 1894-Mar. 1895, Ed. Palmer 551 (F, G, US, C, M).

VERA CRUZ: near Tantoyuca, prov. Huasteca, 1858, Ervendberg 148 (G); sandy soil by Rio de Santa Maria, Zacuapan, Aug. 1906, Purpus 2009 (G); Vera Cruz, 12 March 1910, Orcutt 2999 (F, M); in dry sunny places near fences, 17 March 1857, Mohr (US).

OAXACA: Tuxtepec, 90 m. alt., 24 Aug. 1895, L. C. Smith 647 (G).

CAMPECHE: Canasayal, 20 m. above Chanpotan River, 12 Dec. 1900, Goldman 458 (F).

Yucatan: Izamal, coll. of 1888, Gaumer (F); Merida, 24 Nov. 1864, Schott 22, 23 (F); Buena Vista, 21 June 1892, Gaumer (F); open places, Casa de las Monjas, 20 March 1903, Seler 3995 (G, F); Chicankanab, 1895, Gaumer 394 (F) 364 (US, F, G, M).

COZUMEL: center of Island, 20 Feb. 1899, Millspaugh 1548 (F). CENTRAL AMERICA:

British Honduras: without exact locality, coll. of 1905-1907, *Peck 293* (G).

Guatemala: Puerto Barrios, 2 April 1922, Greenman 5978 (M); Puerto Barrios, Dept. de Izabal, sea level, 2-6 June 1922, Standley 24795 (US); Quirigua, Dept. de Izabal, 75-225 m. alt., 15-31 May 1922, Standley 23762 (US); clear places, Chama, Alta Verapaz, 270 m. alt., 8 June 1920, Johnson 211 (US); eastern Verapaz and Chiquimla, 1885, Watson 346, 3732 (G); Santa Barbara, Dept. of Solola, 415 m. alt., Aug. 1891, Shannon 247 (US); Santa Rosa, 900 m. alt., July 1892, Heyde & Lux 3017 (G).

EL SALVADOR: Ahuachapan, Dept. of Ahuachapan, 800–1000 m. alt., 9–27 Jan. 1922, Standley 19749 (US); vicinity of Sonsonate, Dept. de Sonsonate, 220–300 m. alt., 18–27 March 1922, Standley 22003 (US); Santa Emilia, Sonsonate, 135 m. alt., 22–25 March 1922, Standley 22097 (US); Acajutla, Sonsonate, 30 m. or less alt., 20 March 1922, Standley 21905 (US); Ateos, Dept. de la Libertad, 17 April 1922, Standley 23339 (US); San Salvador, April 1905, Velasco 8854 (G); dry thicket, San Miguel, Dept. de San Miguel, 110 m. alt., 24–27 Feb. 1922, Standley 21114 (US).

NICARAGUA: open ground near Chinandega, 21 Jan. 1903, Baker 754 (US); Granada, 25 Feb. 1903, Baker 166 (G, M).

Costa Rica: along railroad tracks near Moin Junction, 1 Sept. 1919, Rowlee 505 (US); Hacienda de Guacimo, date and collector's name lacking, United Fruit Co. 71 (US).

Panama: vicinity of Cristobal, Colon, 5 Jan.-22 Feb. 1923, Broadway 67 (G, US); Bocas del Toro, 6 Feb. 1921, Carleton 147 (G).

# WEST INDIES:

Bahamas: Governors Harbor, Eleuthera, 14 Dec. 1890, Hitchcock (F, M); Governors Harbor, Eleuthera, 19–20 Feb. 1907, Britton & Millspaugh 5534 (F); Nassau, New Providence, 6 Jan. 1903, Curtis 24 (F, G, US, M); waste places, Nassau, New Providence, 9 Sept. 1904, Britton & Brace 655 (F); Nassau, New Providence, 13 Jan. 1905, Wight 40 (F, G); New Providence, 15 July, 1879, Brace 23 (F); coppice, near Nicholl's Town, northern section of Andros, 4–5 Feb. 1910, Small & Carter 8928 (F); open ground near lighthouse, Watling Island, 13 March 1907, Britton & Millspaugh 6616 (F); sandy soil between dunes, Parrot Cay, Caicos Group, 3 March 1911, Millspaugh 9196 (F); Inagua, 3 Dec. 1890, Hitchcock (F, M).

Cuba: in thickets near trail, Sierra Mendoza, Prov. of Pinar del Rio, 25 Dec. 1911, Shafer 11144 (US, F); Santiago, Prov. of Havana, 10 April 1904, Van Hermann 113 (F); Cienfuegos, Prov. Santa Clara, 24 June 1895, Combs 220 (F, C, G, M); Ingenio Soledad, Cienfuegos, Prov. Santa Clara, 24 Jan. 1903, Pringle 9 (G, US); waste places, La Gloria, Camaguey, 30 Jan. 1909, Shafer 128 (F); forests about Paso Estancia, Oriente, 27 April 1909, Shafer 1576 (US); San Juan Hill, Santiago, 2 Feb. 1899,

Millspaugh 1043, 1048 (F); Santiago, 15-18 Feb. 1902, Pollard & Palmer 329 in part (F).

Haiti: vicinity of Etroite, Gonave Island, 15–21 March 1920, Leonard 3302 (US); vicinity of Pikmi, Gonave Island, 5–9 July 1920, Leonard 5124, 5229 (G, US); Guayubin, Monte Cristi, 100 m. or less alt., 13–21 Feb. 1921, Abbott 1026 (US); waste and cultivated ground, Anse Gallette, Gonave Island, 3–14 March 1920, Leonard 3101 (US); St. Marc, sea level, 25–28 Feb. 1920, Leonard 2943 (G, US); Puerto Frances, Samana Peninsula, sea level to 200 m. alt., 28–29 March 1921, Abbott 1200 (US); Samana Peninsula, sea level to 200 m. alt., 30 Dec. 1920, Abbott 498 (US); Hispaniola, Puerto Plata, 26 April 1906, Raunkiaer 869 (US); Haina, July 1921, Faris 303 (US); railroad, Pimental, Prov. Pacificador, 20–25 Jan. 1921, Abbott 641, 648 (US); sandy loam, open hilltops, July 1921, Faris 324 (US); Barahona, April 1911, Fuertes 886 (US); locality lacking, Jan.-March 1871, Wright, Parry & Brummel 354 (US).

Porto Rico: Santurce, 8 Nov. 1899, Goll 68 (US); Camuy, 21 June 1901, Underwood & Griggs 201a (US); Santa Anna, 9 Nov. 1899, Goll 150 (US); Cantano, 6-11 Jan. 1899, Millspaugh 337 (F); near hot springs, Coama, 1 July 1901, Underwood & Griggs 529 (US); open fields, Coama Springs, 22 Nov. 1899, Goll 675 (US); Mayaguez, 11 Jan. 1884, Sintenis 157 (G); Adjuntas Road, seven miles from Ponce, 2 Dec. 1902, Heller 6181 (G, F, M); roadside near Izabel Segunda, Vieques Island, 24-27 Jan. 1914, Shafer 2425 (US); locality lacking, 1899, Heller 149 (F).

Grand Cayman: Spot Bay, 13-14 Feb. 1899, Millspaugh 1286 (F); Jan. 1890, Hitchcock (M).

Jamaica: rocky bank at roadside in vicinity of Montego Bay, 28-30 March 1920, Maxon & Killip 1605 (US, G); woods near Port Antonio, 29 June 1897, Fredholm 3081 (US); Port Antonio, 28 Jan.-6 Feb. 1899, Millspaugh 921 (F); Porus, Jan. 1892, Lloyd 1104 (F, M); Ferry River on Spanish Town road, sea level, 24 May 1904, Maxon 2180 (US); along railroad between Kingston and Gregory Park, sea level, 22 Feb. 1920, Maxon & Killip 313 (US); dryish situation near Kingston, 28 April 1903, Maxon 1657 (US); streets in Kingston, 9 Dec. 1890, Hitchcock (M); Public Gardens, Hope Grounds, Kingston, 210 m. alt., 13 Nov. 1914,

Harris 11796 (F, US, C, M); open railroad embankment at St. Margaret's Bay, Nov. 1900, Millspaugh 1910 (F); on Windward Road, 27 Aug. 1902, Harris (F); King's House, Campbell 6785 (F); locality lacking, Dec. 1869, Alexander (F, US, G).

St. Jan: rocky hillside, Lamosure, 10-12 Feb. 1913, Britton & Shafer 507 (US).

Lesser Antilles: near Bassin Yard, St. Croix, Dec. 1895, Ricksecker 167 (F, G, US, M); near Bassin Yard, St. Croix, 17 Feb. 1897, Ricksecker 131 (F, US, M); St. Thomas, Dec. 1886, Eggers (F); among fields in St. Thomas, Jan. 1887, Eggers 26 (US); St. Thomas, Dec. 1880, Eggers 365 (G); St. Thomas, Ehrenberg 107 (M); St. Thomas, Krebs (F); roadside at Kinsale, Montserrat, 22 Jan. 1907, Shafer 138 (F); Martinique, date lacking, Sieber 316 (M); Barbados, coll. of 1900, Botanic Station Herbarium 139 (F, G, US); roadside ditch, St. Vincents, March 1890, Smith 714 (G); in cocoa fields, Grenada, Oct. 1904, Broadway (US, M); Government House, Tobago, 19 Nov. 1913, Broadway 4845 (US); bank of saddle road, Trinidad, 28 Feb. 1920, Britton & Hazen 162 (US, G).

# SOUTH AMERICA:

COLOMBIA: Santa Marta, 30 m. alt., Nov. 1898–1901, Smith 1465 (F, G, US, M); Santa Marta, 75 m. alt., Nov. 1898–1901, Smith 545 (F, G, US, M); Boco Verde, on Rio Sinu, Cacaolate, Dept. of Bolivar, 90–120 m. alt., 13–14 Feb. 1918, Pennell 4198 (US, G).

VENEZUELA: Margarita Island, 9 July 1901, Miller & Johnston 96 (F, G, US, M); Sacuapana, April 1896, Rusby & Squires 306 (F, G, US, M); near colony of Tovar, 16 Aug. 1855, Fendler 912 (G).

British Guiana: Promenade Gardens, Georgetown, 30 Oct.-1 Nov. 1919, *Hitchcock 16593* (US, G); weed in the field, Mahaica, on coast, 20 miles east of Georgetown, 15 Nov. 1919, *Hitchcock 16773* (G, US).

French Guiana: grassy places, Cayenne, 2 May 1921, Broadway 89 (US, G).

Ecuador: Caraques Bay, 17 June 1923, Anthony and Tate 118 (US); occasional in shady places around 300 m. alt., on Charles

Island, Galapagos Islands, 28 Feb. 1905, Stewart 3312 (G, US, M).

PERU: sandy roadside, La Merced, 600 m. alt., 10-24 Aug. 1923, *Macbride 5296* (F); locality and date lacking, *Ruiz 4785* (US).

Bolivia: junction of the rivers Beni and Madre de Dios, Aug. 1886, Rusby 1784 (F, US, G); Guanai-Tipuani, Apr.-June 1892, Bang 1375 (F, G, US, M).

# 7. Priva armata Watson, Proc. Am. Acad. 25: 160. 1890.

A low slender herb, 3 dm. or less high, much branched from the base, lower branches often arcuate-ascending; stem slender, square, more or less pubescent; leaves sessile, ovate, 10–15 mm. long, 6–7 mm. broad, irregularly toothed, acute at the apex, pubescent; spikes few-flowered, short; bracts broadly ovate, spatulate, rough, hispid, 4–5 mm. long; calyx tubular, finely pubescent, accrescent, at maturity subglobose, 8–10 mm. long, loosely inclosing the fruit, thin, membranaceous; fruit hard, consisting of two bilocular cocci, dorsal surface covered with stout straight spines, commissural surface flat.

Distribution: near Monterey, Mexico.

Specimens examined:

MEXICO: Valley of Monterey, 7 July 1889, Pringle 1931 (G, TYPE, F); Valley of Monterey, 18 July 1889, Pringle 2674 (M, C).

8. Priva cuneato-ovata (Cav.) Rusby, Bull. Torr. Bot. Club 27: 80. 1900.

Castelia cuneato-ovata Cav. Anal. Cienc. Nat. Madrid 3: 134. 1801; Ic. and Des. Pl. 6: 60. t. 583. 1801.

Priva laevis Juss. Ann. Mus. Paris 7: 70. 1806; Pers. Syn. Pl.2: 139. 1807; Walpers, Rep. 4: 36. 1844–1847.

Verbena tuberosa R. Graham, Edinb. N. Phil. Jour. 29: 174. 1840.

Priva orchioides Walpers, Rep. 4: 36. 1844-1847.

Bouchea copiapensis Gay, Hist. Nat. Chile 5: 26, Atlas 1, pl. 55. 1849.

Stem simple or branched, 3-4.5 dm. high, 4-angled, lower branches often arcuate-ascending, glabrous or very slightly

pubescent; leaves petiolate to nearly sessile, ovate to subrotund, 3-8 cm. long, 1-4 cm. broad, coarsely mucronate-serrate to crenate, acute to obtuse at the apex, attenuate at the base into a petiole, glabrous or somewhat puberulent; racemes terminal, flowers distant, opposite or distinctly verticillate, shortly pedicellate; bracts lanceolate, acuminate, 4-7 mm. long; calyx pubescent, 10-12 mm. long, folded, with long acuminate lobes, tips of calyx lobes involute, hyaline-margined, more or less contorted over the fruit at maturity; corolla bluish-red, tube pubescent, spread of limb approximately equaling the length of the calyx-tube; fruit included within the persistent calyx, ovate, 4-5 mm. long, 2-3 mm. wide, splitting at maturity into two bilocular cocci, dorsal surface of individual coccus convex, somewhat longitudinally ridged, commissural surface flat.

Distribution: moist places, Argentina and Chile.

Specimens examined:

SOUTH AMERICA:

ARGENTINA: Territorio del Chaco, 9 April 1918, Jörgensen 2480 (G, US, M); Dept. Andalgala, Prov. de Catamarca, 16 Sept. 1918, Jörgensen 1022 (G, US, M); Cordoba, Nov. 1892, Kuntze (F).

CHILE: along irrigating ditches and in moist places, Tacna Arica region, 20 April 1922, Shepard 269 (US); Vallenar, Prov. Atacoma, 300 m. alt., 1 Feb. 1923, Werdermann 137 (M); Santiago, Jan. 1919, Bro. Claude-Joseph 735 (US); Santiago, Jan. 1919, Bro. Claude-Joseph 804 (US); without exact locality, Oct. 1914, Buchtien 4381 (US).

9. Priva rhinanthifolia (Mart. & Gal.) Robinson, n. comb. Verbena rhinanthifolia Mart. & Gal. in Bull. Acad. Brux. 11<sup>2</sup>: 323. 1844.

Priva tuberosa Watson, Proc. Am. Acad. 18: 135. 1883.

Roots tuberous; stems branched at the base, erect or decumbent, 4-angled, hairy, 5-30 cm. long; leaves sessile, oblong, 1.5-4 cm. long, 0.5-1 cm. broad, coarsely toothed, especially toward the apex, somewhat narrowed at the base, more or less strigosely pubescent on the upper surface, sometimes uniformly, often in patches, pubescence prominent on the under surface, especially along the midrib and veins; racemes extremely short and few-

flowered; bracts lanceolate, pubescent, 4–5 mm. long; calyx unequally and deeply 5-lobed, conspicuously pubescent, becoming 8–9 mm. long, somewhat accrescent, more or less constricted over the cocci at maturity; corolla exceeding the calyx-tube, pubescent; fruit somewhat globose, consisting of two bilocular cocci which are irregularly and coarsely reticulated, not spiny.

Distribution: central and southern Mexico.

Specimens examined:

Mexico:

CHIHUAHUA: Parral, 1800 m. alt., 19 Sept. 1898, Goldman 103 (US); oak woods and plains near Cosihuiriachic, 27 Aug. 1887, Pringle 1549 (G, C); under oaks, hills near Cosihuiriachic, 19 Sept. 1888, Pringle 3057 (F).

Durango: Otinapa, 25 July-5 Aug. 1906, Ed. Palmer 396 (M, G); eity of Durango, 1 Aug. 1898, Nelson 4635 (F, G, US); grassy sides of ravines at Santiago Papasquiara, Apr.-Aug. 1896, Ed. Palmer 424 (F, G, M).

SAN Luis Potosi: in the mountains near San Miguelito, 1876, Schaffner 713 (C).

Hidalgo: calcareous soil near Tula, 2640 m. alt., 25 July 1898, Pringle 7586 (G, F).

Jalisco: road between Huejuquilla and Mesquite, 25 Aug. 1897, Rose 3576 (US).

Mexico: Teoloyucan, 11 Aug. 1913, Salazar (US); vicinity of Tlalnepantla, 6 July 1905, Rose 8422 (US); near Guadalupe, Valley of Mexico, 1905, Rose 8511 (US); San Angel, Valley of Mexico, 1865–1866, Bourgeau 357 (G, US); San Angel, Valley of Mexico, 15 Aug. 1905, Rose 9496 (US).

MICHOACAN: Punguato, 2100 m. alt., 20 June 1912, Arsène 8296 (US); grassy hills near Patzcuaro, 19 July 1892, Pringle 4147 (C, F, G, M); Lama Santa Maria, 1950 m. alt., 14 June 1909, Ars'ne 3499 (US).

10. Priva aspera HBK. Nov. Gen. & Sp. 2: 278. 1817; Walpers, Rep. 4: 34. 1844; Schauer in DC. Prodr. 11: 534. 1847; Engl. & Prantl, Nat. Pflanzenfam. 4: Abt. 3a, 155. 1895.

Priva trachelioides Mart. & Gal. in Bull. Acad. Brux. 11<sup>2</sup>: 324. 1844.

Priva Orizabae Watson, Proc. Am. Acad. 23: 282. 1888.

Stem erect, 12–18 dm. high, branched; branches four-sided, striate, pubescent; leaves petiolate, ovate, 6–20 cm. long, 3–10 cm. wide, crenate, acuminate, acute at the base, scabrous above, pale beneath, reticulately veined, nerves and veins rather prominent beneath; racemes terminal, solitary or in threes, 15–45 cm. long; bracts lanceolate, 1–2 mm. long; flowers solitary, distant; pedicels stout, 1.5–2 mm. long; calyx tubular in flower, lobes involute, hyaline-margined, sparsely pubescent, in some instances pubescent with both straight and uncinate hairs, globose in fruit, sulcate; corolla bilabiate, slightly exceeding the calyx, tube pubescent, throat hairy; fruit inclosed in the persistent calyx, erect, consisting of two 1-celled cocci, dorsal surface convex, reticulately ridged, without spines, commissural surface oblique, excavated.

Distribution: Mexico and Central America.

Specimens examined:

MEXICO:

CHIHUAHUA: Santa Eulalia Mts., 8 Sept. 1885, Pringle 287 (G, C, F, US, M); Sierra Madres near Seven Star Mine, 2100 m. alt., 15 Sept. 1899, Townsend & Barber 422 (G, F, US, M); Sierra Madre Mts. in Guayanopa Canyon, 1080 m. alt., 24 Sept. 1903, Jones 7323 (G).

SAN Luis Potosi: near San Luis Potosi, 1800-2400 m. alt., 1787, Parry & Palmer 713 in part (M).

SINALOA: Santa Lucia, Sept. 1919, Dehesa 1644 (US); in thickets along the Rio Fuerte, near San Blas, 24 March 191-, Rose 13371 (US).

Tepic Territory: 5 Jan.-6 Feb. 1892, Ed. Palmer 1999 (G, F, US).

Jalisco: Guadalajara, July-Oct. 1886, Ed. Palmer 500 (G, US).

Vera Cruz: moist rocky slopes, Zacuapan, Sept. 1906, Purpus 1921 (F, G, M); Zacuapan, Sept. 1917, Purpus 8054 (G, US, M); Mt. Orizaba, 1865–1866, Bourgeau 2950 (G, US); near Orizaba, 1200 m. alt., July 1891, Seaton 465 (G); Orizaba, 25 Sept. 1865–1866, Bourgeau 3118 (G); Orizaba, without date, Botteri 593 (G). Michoacan: vicinity of Morelia: northwest of Punguata, 570

m. alt., Sept. 1900, Arsène (US, M); Rincon, 1950 m. alt., 21 Aug. 1922, Arsène 8696 (F, US, M); Rincon, 1900 m. alt., 8 Sept. 1910, Arsène 5292 (G, US, M); Rincon, 1850 m. alt., 19 Sept. 1909, Arsène 2545 (US); Rincon, 1950 m. alt., 25 July 1909, Arsène 2796 (G, M); 2000 m. alt., 9 Nov. 1909, Arsène (US); Morelia, Apr. 1909, Arsène 45 (F).

OAXACA: Rancho de Coldevin, 1650 m. alt., 10 Sept. 1894, L. G. Smith 160 (G).

# CENTRAL AMERICA:

GUATEMALA: Coban, Dept. Alta Verapaz, 1350 m. alt., Sept. 1907, von Türckheim 1628 (G); coll. of 1892, Heyde 206 (US).

COSTA RICA: locality and date lacking, Kuntze (F).

# 11. Priva mexicana (L.) Pers. Syn. Pl. 2: 139. 1807.

Verbena mexicana L. Syst. 66. 1784; Willd. Sp. Pl. 1: 116. 1797.

Zapania mexicana Lam. Ill. Gen. 1: t. 17, f. 1. 1823; Poir. Dict. 8: 845. 1808.

Blairia mexicana Gaertn. Fruct. 1: 265. t. 56. 1787.

Priva hispida Juss. Ann. Mus. Paris 7: 70. 1806; Walpers, Rep. 4: 34. 1844; Schauer in DC. Prodr. 11: 534. 1847.

(?) Priva crenata Schrad. Ind. Sem. Hort. Götting, 1831; Linnaea 8, Litteratur-Bericht: 24. 1833.

Stem erect, simple or branched, quadrangular, striate, 3-12 dm. high, more or less pubescent; leaves short-petiolate or sessile, ovate, subcordate, 2-8 cm. long, 1-5 cm. broad, somewhat crenate-dentate, acute at the apex, subcordate at the base, strigosely pubescent on the upper surface, pale beneath; racemes terminal or axillary, pedunculate, erect or subflexuous, 5-30 cm. long; bracts lanceolate, usually longer than the pedicels; pedicels very minute, 0.5-1 mm. long; calyx in flower cylindrical, densely uncinate-hispid, fruiting calyx globose, close-fitting, connivent at orifice, 2-parted, splitting at maturity of fruit; corolla usually twice the length of the calyx, lilac; fruit consisting of 2 unilocular cocci, reflexed, convex on the dorsal surface, reticulately ridged, without spines, commissural surface somewhat concave on either side of a persistent longitudinal median ridge, smooth, not margined.

Distribution: Mexico.

Specimens examined:

. Сніниания: shaded ravines and mesas near Cosihuiriachic, 28 Aug. 1887, Pringle 1354 (G, US, F).

COAHUILA: Saltillo, Sept. 1898, Ed. Palmer 281 (G, US, M).

Durango: near the city of Durango, Apr.-Nov. 1896, Ed. Palmer 578 (G, F, US, M); Ramos Inde, 11-14 Aug. 1898, Nelson 4709 (US).

Nuevo Leon: Monterey, 800 m. alt., Aug. 1911, Abbon 6178 (US).

SAN LUIS POTOSI: Minas de San Rafael, July 1911, Purpus 5518 (G, F, US, M); region of San Luis Potosi, 1800–2400 m. alt., 1878, Parry & Palmer 713 (G, F, M, US).

HIDALGO: Zimapan (fide Hemsley), date lacking, Coulter 1141 (G); near Tequixquiac, 30 Aug. 1903, Rose 6642 (US); hills near El Salta, 2100 m. alt., 17 Sept. 1901, Pringle 9287 (G, F, M).

Mexico: Mexico City, Apr.-Nov. 1896, Ed. Palmer 578 (M); Mixcoac, Federal District, 11 Aug. 1913, Arsène 8808 (US); Mixcoac, Federal District, 11 Aug. 1913, Arsène 8508 (F, M); Tlalpam, Federal District, 1910, Orcutt 3628 (US, F, M); Valley of Mexico, 18 June 1865, Bourgeau 359 (G); Valley of Mexico, 1875, Schaffner 425 (G).

Puebla: vicinity of Puebla: 2170-2270 m. alt., 27 Oct. 1907, Arsène 1195 (US); Mayorazzo sier l'Atayac, 2120 m. alt., 4 July 1907, Arsène 1340 (US); Cerro Tepaxuchil, 2330 m. alt., 11 July 1907, Arsène 10207 (US); Cerro Tepaxuchil, 2330 m. alt., 14 Nov. 1908, Arsène 7054 (US); Santa Barbara, 2150 m. alt., 20 June 1910, Nicolas & Arsène 5280 (US, M); San Luis Tultitlanapa, July 1908, Purpus 3524 (F).

OAXACA: Valley of Oaxaca, 1550 m. alt., 21 July 1897, Gonzalez 290 (G); near Cuicatlan, 750-1200 m. alt., 24 Oct. 1894, Nelson 1822 (US, G).

MICHOACAN: vicinity of Morelia: Lorna Santa Maria, 1950 m. alt., 28 Aug. 1910, Arsène (US, M); Jaripeo, 2100 m. alt., 13 July 1911, Arsène (G, US, M).

# LIST OF EXSICCATAE

The distribution numbers are printed in *italics*. Unnumbered collections are indicated by a dash. The number in parenthesis is the species number used in this revision.

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Millspaugh, C. F. & C. M. 9196 (6).

Nelson, E. W. 1822, 4709 (11); 4635

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Montes, M. N. 389 (6).

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# EXPLANATION OF PLATE

# PLATE 1

Geographical distribution of the genus Priva.

# WESTERN HEMISPHERE

= Priva lappulacea.
= Priva mexicana.
= Priva aspera.
= Priva cuneato-ovata.
= Priva rhinanthifolia.
= Priva armata (in Mexico).
= Priva domingensis (in West Indies).
= Priva portoricensis (in West Indies).
= Priva bahiensis (in South America).
EASTERN HEMISPHERE.
= Priva leptostachya.
= Priva Curtisiae.

## EXPLANATION OF PLATE

PLATE 2

Priva Curtisiae Kobuski

British East Africa

From the type specimen, Curtis No. 499, in the Gray Herbarium of Harvard University.



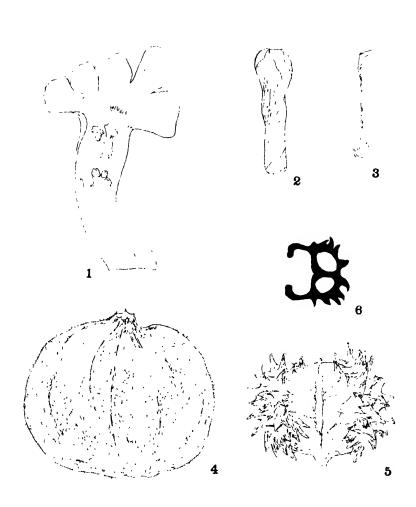
KOBUSKI REVISION OF GENUS PRIVA

## EXPLANATION OF PLATE PLATE 3

#### Priva Curtisiae Kobuski

- Fig. 1. Open corolla,  $\times$  6. Fig. 2. Corolla in bud,  $\times$  6.

- Fig. 3. Pistil, × 6.
  Fig. 4. Mature calyx, × 6.
  Fig. 5. Mature fruit, × 6.
- Fig. 6. Cross-section of coccus,  $\times$  6.

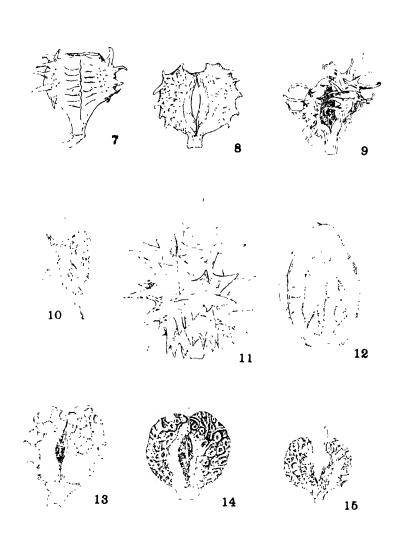


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### EXPLANATION OF PLATE

#### PLATE 4

- Fig. 7. Fruit of Priva portoricensis Urban,  $\times$  6.
- Fig. 8. Fruit of Priva leptostachya Juss., × 6.
- Fig. 9. Fruit of Priva bahiensis DC., × 6.
- Fig. 10. Fruit of Priva lappulacea (L.) Pers., × 6.
- Fig. 11. Fruit of Priva armata Watson, × 6.
- Fig. 12. Fruit of Priva cuneato-ovata (Cav.) Rusby, × 6.
- Fig. 13. Fruit of Priva rhinanthifolia (Mart. & Gal.) Robinson, × 6.
- Fig. 14. Fruit of Priva aspera HBK., × 6.
- Fig. 15. Fruit of Priva mexicana (L.) Pers., × 6.

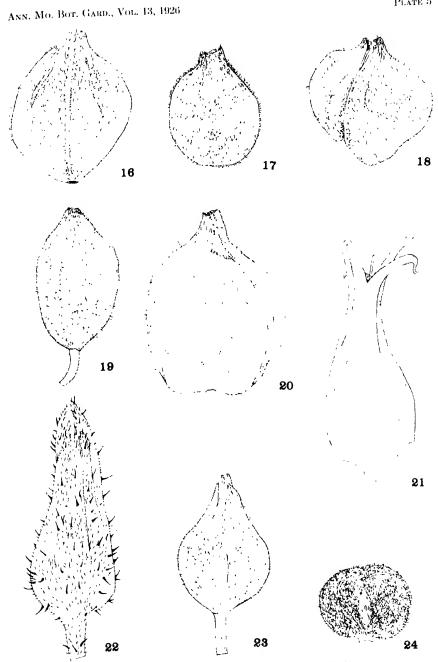


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### EXPLANATION OF PLATE

#### PLATE 5

- Fig. 16. Mature calyx of Priva portoricensis Urban, × 6.
- Fig. 17. Mature calvx of Priva leptostachya Juss., × 6.
- Fig. 18. Mature calyx of Priva bahiensis DC., × 6.
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- Fig. 20. Mature calyx of Priva armata Watson,  $\times$  6.
- Fig. 21. Mature calyx of Priva cuneato-ovata (Cav.) Rusby, × 6.
- Fig. 22. Mature calyx of Priva rhinanthifolia (Mart. & Gal.) Robinson, × 6.
- Fig. 23. Mature calyx of Priva aspera HBK., × 6.
- Fig. 24. Mature calyx of Priva mexicana (L.) Pers., × 6.



KOBUSKI REVISION OF GENUS PRIVA

#### STUDIES ON SOUTH AMERICAN LABIATAE. II<sup>1</sup>

#### Synopsis of the Genus Sphacele

#### CARL CLAWSON EPLING

Instructor in Botany, University of California, Southern Branch Formerly Rufus J. Lackland Research Fellow in the Henry Shaw School of Botany of Washington University

#### SPHACELE

SPHACELE Bentham in Edwards' Bot. Reg. sub. *pl. 1289*. 1829; Lab. Gen. et Sp. 567. 1834; et in DC. Prodr. 12: 254. 1848, nom. conservandum.

Algue Laguen Feuillée, Hist. Pl. Medecin. 4. 1725.

Alguelaguen Adanson, Fam. 2: 505. 1763.

Dracocephalum Balbis in Mem. Acad. Sci. de Turin 12<sup>1</sup>: 345. pl. 7. 1802-3 (non L.).

Phytoxis Molina, Sagga sulla Storia Nat. del Chili, ed. 2, 145, 290. 1810.

Sideritis Kunth in Humboldt & Bonpland, Nov. Gen. et Sp. Pl. 2: 306. 1817 (non L.).

Phytoxys Sprengel, Syst. 2: 676. 1825.

Alguelagum Kuntze, Rev. Gen. 2: 511. 1891.

Frutices suffruticesve habitu foliisque fere Salviae, his saepius bullato-rugosis, subtus tomentosis; floribus in paniculis ad ramorum extremitates dispositis, rarius solitariis, saepius in verticillastris densioribus confertis; calycibus florentibus campanulatis vel tubuloso-campanulatis, subbilabiatis, dentibus lanceolato-subulatis, tubo frequenter aequilongis vel brevioribus, maturis nunc valde nunc paulo auctis, saepius chartaceis, frequenter inflatis; corollae tubo rarius calyci duplo longiore, saepius breviter exserto, superne gradatim ampliato, intus rarius omnino annulato, nectarostegio maximam partem e pilis diffusis areolam ad staminum bases facientibus consistente, limbo leniter bilabiato,

Issued May 8, 1926.

<sup>&</sup>lt;sup>1</sup> Part I of Studies on South American Labiatae published in Ann. Mo. Bot. Gard. 12: 107-132. 1925.

lobis subaequalibus, labioli medio majore; staminibus inclusis, loculis subparallelis; stylo subaequaliter bifido, corollam subaequante; nuculis atris, laevibus, obovatis, hilo obscuro; gynobase nullomodo aucto.

The term "Algue Laguen" was employed by Feuillée as a common name, being a transcription from the Araucanian dialect (see following under Sphacele chamaedryoides). name was compiled by Adanson unchanged, with a hardly sufficient diagnosis, and latinized by Kuntze. Phytoxys of Sprengel was a compilation of Molina's name with slightly different spelling. The genus was not clearly understood until outlined by Bentham.

Sphacele chamaedryoides (Balbis) Briquet (S. campanulata Bentham) is herein considered the type species, since it is the species longest known and the first to be described, although not under the name Sphacele. Since Bentham did not employ the more recent concept of a type it is impossible to assign the role to any of the species of the genus as understood by him. In the preliminary synopsis in the 'Botanical Register,' Sphacele Lindleyi (Stachus Salviae Lindl.) was the first mentioned. In the monograph Sphacele salviaefolia (Sideritis salviaefolia Kunth) was the first described. In the account of the genus prepared for De Candolle's 'Prodromus' several newly described species were added, the entire arrangement altered, and Sphacele speciosa, which had appeared as the last species in the monograph, was here placed first, together with allied species.

#### CONSPECTUS SPECIERUM

A. Flores in axillis foliorum vel bracteorum solitarii.

- b. Flores in axillis bracteorum parvorum solitarii. 1. Folia anguste oblonga vel oblanceolata, in basi extenuata 2. Folia lanceolata vel ovato-lanceolata, in basi truncata vel sub-B. Flores 2-numerosi in verticillastris dispositi. a. Flores plerumque tres in verticillastris; racema patula, internodiis
  - sat longis.
    - 1. Bractea foliosa, calycibus subaequilonga; calyx maturus floccoso
      - a. Calyx tubulosus; corollae tubum patenter tubulosum ...... 4. S. Salviae

β. Calyx subcampanulatus; corollae tubum late tubuloso- campanulatum
2. Bractea parva, pedicellis subaequilonga; calyx maturus glabrus vel parce hirsutus.
α. Folia ovato-trigona vel oblonga, in basi hastata.
I. Folia subtus dense tomentosa; corolla 2–2.5 cm. longa
II. Folia subtus praecipue ad venas crispuli-tomentosa;
corolla 1.5 cm. longa
β. Folia oblonga vel lanceolata, in basi rotundato-angustata.
I. Folia glabra, petiolis nec alatis
b. Flores infra plerumque 6-9, supra 3-4 in verticillastris; in paniculis
densis vel spicato-interruptis.
<ol> <li>Flores in paniculis densis, frequenter laxis, vix interruptis, rarius subglobosis.</li> </ol>
a. Suffrutex floribus in spicis oblongis vel subglobosis saepius
pedunculatis confertis
β. Frutices floribus in paniculis amplis dispositis.
I. Corolla 6-12 mm. longa.
Folia bullato-rugosa, in basi subtruncata.
11. S. salviaefolia
II. Corolla plerumque longitudine minus quam 6 mm.
* Bractea acutiuscula, calyces florentes super-
antia.
† Folia in basi rotundato-angustata vel
extenuata.
‡ Calycis dentes maturi lanceolato-
acuminati.
Folia in basi rotundato-an-
gustata, non extenuata;
calycis dentes 2.5-3 mm.
longi
Folia in basi extenuata; caly-
cis dentes 1.5–2 mm. longi
## Calycis dentes maturi ovato-trigoni,
acuti vel obtusi13. S. aurifera †† Folia in basi patenter truncata vel cor-
data
** Bractea obtusiuscula, saepius rotundata calyces
florentes vix aequantia.
† Folia in basi truncata vel cordata.
‡ Folia oblonga, in basi truncata saepe
subhastata.
Panicula canescenti-tomen-
tosa; folia truncata, apice
obtusa19. S. intermedia

Panicula pubescentia; folia truncata, subhastata, apice acuta vel lenissime acumin- ata18. S. conferta
II Folia ovata vel ovato-elliptica, in
basi profundo- vel truncato-
cordata.
Folia ovata, profunde cordata
Folia ovato-elliptica, in basi
anguste truncato-cordata
†† Folia in basi angustata, anguste oblonga
2. Flores in panicula submoniliforma, verticillastris inter se .5-1
cm. distantibus.
α. Folia in basi truncato-extenuata20. S. Sprucei
β. Folia in basi rotundata vel extenuata.
I. Folia lanceolata, superficie $10-12 \times 4-6$ cm.
15. S. acuminata
II. Folia oblongo-elliptica superficie vel $10-17 \times 2.5-4$
cm. vel $5-8 \times .7-1.5$ cm.
* Folia superficie $10-17 \times 2.5-4$ cm.
** Folia superficie 5-8 × .7-1.5 cm

1. Sphacele tomentosa Benth. Lab. Gen. et Sp. 569. 1834. Alguelagum tomentosum Kuntze, Rev. Gen. 2: 511. 1891.

Suffrutex diffusus, humilis et prostratus, ramis teretibus, glabris, cortice discedente, ramulis obtuse quadratis, tomentosis; foliis 10-15 mm. longis, internodia maximam partem superantibus, oblongis, obtusis, in basi rotundato-subhastatis, margine convexiuscula, crenato-dentata, crenis obtusis, pagina superiore hispidula, viride, rugosa, inferiore cano-tomentosa, venis prominulis, pedicellis 5-8 mm. longis elatis; floribus in axillis foliorum superiorum solitariis, oppositis; calveibus florentibus 5 mm. longis, tubuloso-campanulatis, bilabiatis, hispido-tomentellis, dentibus aequilongis, tubo paulo brevioribus, acuminatis, fructiferibus 9-10 mm. longis, dentibus recurvis, posticis tribus sinu rotundato latiore ab anticis separatis; corollis 6 mm. longis, tubo 3.5 mm. longo, nectarostegio interrupte annulato, intus ad tubi basim e pilis consistente, limbo bilabiato, labro 1.5 mm. alto, emarginato, labiolo trifido, lobis lateralibus quam labro brevioribus, medio rotundato subduplo longiore; staminibus ad tubi medium in-

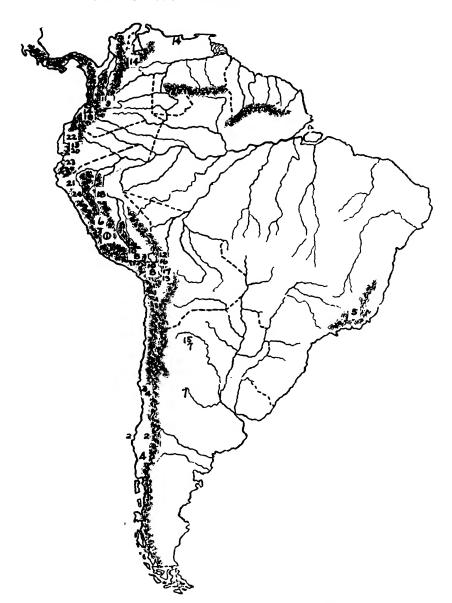


Fig. 1. Map showing distribution of the species of Sphacele, the numbers being those assigned to the species described in this paper.

sertis, didymis; stylo tubo aequilongo, superne dilatato; nuculis non visis.

Specimens examined:

PERU: no data given, Dombey 278 (GH, type collection). The locality recorded by Bentham is Cheuchin.

2. Sphacele chamaedryoides Briq. in Engl. & Prantl, Nat. Pflanzenfam., IV. Abt. 3 a, 291. 1897.

Dracocephalum chamoedryoides Balbis in Mem. Acad. Sci. de Turin 12: 345. pl. 7. 1802-3.

Phytoxis sideritifolia Molina, Sagga sulla Storia Nat. del Chili, ed. 2, 145. 1810.

Phytoxis acidissima Molina, loc. cit. 290. 1810.

Sphacele campanulata Benth. in Edwards' Bot. Reg. 15: sub. pl. 1289. 1829; Lab. Gen. et Sp. 569. 1834; in DC. Prodr. 12: 255. 1848.

Alguelagum chilense Kuntze, Rev. Gen. 2: 511. 1891.

Sphacele chilensis Briq. Bull. de l'Herb. Boiss. 4: 805. 1896

Frutex subrigidus, ramosus foliosusque circa 1 m. altitudine. ramis divaricatis, teretibus, glabris, cortice discedente, ramulis gracilibus superne puberulis, mox glabriusculis, teretibus, internodiis minus quam foliorum longitudine; foliis saepe in axillis fasciculatis, 1.5-4 cm. longis, .5-1 cm. latis, oblongis oblanceolatisque, apice obtusioribus, in basi saepius in petiolum brevem attenuatis, margine patenter revoluta, pulchre et regulariter crenata, crenarum culminibus saepius minus quam .5 mm. altis, inter se 1.5-2 mm. distantibus, pagina superiore atroviride, glabra, bullata, inferiore albo-tomentosa, vena media prominentiore, venis lateralibus reticulatis vel obscuris, petiolis 3-5 mm. longis elatis; floribus in panicula foliosa in axillis foliorum vel bracteorum solitatim dispositis, internodiis pedicellis subduplo-triplo longioribus, bracteis inferioribus foliosis 1-1.5 cm. longis, superiore membranaceis, oblongo-lanceolatis, acuminatis, puberulis, maturitate saepe deciduis; calveibus florentibus 8-10 mm. longis, campanulatis, membranaceis, puberulis, tubo 4-5 mm. longo, dentibus subaequilongis, lanceolato-acuminatis, fructiferibus 15-18 mm. longis, dentibus tubum aequantibus. acuminatis vix tamen spinosis, bilabiatis, labia antica longiore, a postica sinibus rotundatis latioribus separatis, corollis 15-18

<sup>&</sup>lt;sup>1</sup> For explanation of abbreviations of herbaria, see Studies on South American Labiatae. I. Ann. Mo. Bot. Gard. 12: 107. 1925.

mm. longis, puberulis, campanulatis, bilabiatis, labri lobis 2–2.5 mm. longis, labioli lateralibus 2–2.5 mm. longis, medio 3.5–4 mm. longo, omnibus rotundatis, nectarostegio annulato supra basim 4 mm. e pilis longis consistente; staminibus didymis, posticis 3–4 mm. longis, supra tubi medium, anticis 8–9 mm. longis, ad tubi medium positis, omnibus e tubo exsertis; stylo 15 mm. longo, exserto; nuculis obovatis, atris, 3 mm. longis.

Specimens examined:

CHILI: Chillan, Dec. 1869, Couthouy (GH); no data, Gay 149 (GH; US; NY); no data, Styles (ASP); no locality stated, Apr. 15, 1868, Shuttleworth (NY); Panguipuilli, Oct. 1923, Bro. Claude-Joseph 2377 (US); Southern Chili, 1828, \*Bertero (MBG); Isle St. Marys, Eights (US); Ercilla, Feb. 1892, Kuntze (US).

The first binomial clearly to be applied to this species was that of Balbis, namely, *Dracocephalum chamoedryoides*. His well-executed plate and the description leave no question as to this, although the author was ignorant of the origin of the seed from which the plants described had been grown.

Previously, however, in the first edition of a work by Molina² but not in the second edition, was described a plant called Rosmarinus chilensis. This plant was referred questionably by Bentham³ to synonymy with his newly described species Sphacele campanulata. Kuntze,⁴ however, referred it here without question and made the new combination Alguelagum chilense. Both authors apparently overlooked the fact that Rosmarinus chilensis was described under the Linnaean class "Diandria," with the corolla as having two stamens, true of Rosmarinus and not true of Sphacele. Since Molina was a careful worker and good observer, as may be learned by a perusal of his work, and since he was undoubtedly familiar with the Mediterranean Rosmarinus, there is no basis for assuming that he described any other than Rosmarinus under that name. This conclusion is further strengthened by the fact that Rosmarinus officinalis is found naturalized

<sup>&</sup>lt;sup>1</sup> Balbis in Mem. Acad. Sci. Turin 12<sup>1</sup>: 345. pl. 7. 1802-3.

<sup>&</sup>lt;sup>a</sup> Molina, I.I. Sagga sulla Storia Naturale del Chili, ed. 1, 309. 1786. (transl. J. D. Brandis), original Italian edition not seen by the author.

<sup>&</sup>lt;sup>2</sup> Bentham, G. Lab. Gen. et Sp., 569, 1834; et in DC. Prodr. 12: 255, 1848.

<sup>&</sup>lt;sup>4</sup> Kuntze, O. Rev. Gen. 2: 511. 1891.

in various Latin-American regions, and in these countries is often grown in gardens, being used as a condiment.

In the second edition of Molina's work<sup>1</sup> the present species was, however, clearly described, this time under the generic name of Phytoxis. Feuillée<sup>2</sup> had previously described the plant by the phrase Algue Laguen Sideritidis folia magno flore subcaeruleo, and Molina, in describing the plant upon which he based not only the species but the genus Phytoxis, made it clearly synonymous with that described by Feuillée and apparently derived his binomial from Feuillée's phrase. It must be remembered that the term "Algue Laguen" was not employed in a generic sense but as a common name, the word "Lahuen" or "Laguen" (a low shrub) having been derived from the Araucanian dialect and applied by Feuillée and others in various combinations, as, for example, "Cachan-lahuen" (Chironia chilensis Willd.). Alguelaguen meant literally "devils shrub." In addition to the generic and specific description of Phytoxis sideritifolia in Italian, on page 145, Molina published a second more complete generic description but less complete specific description of the same plant in the Latin appendix (p. 290) in which all plants discussed in the narrative were arranged according to the Linnaean system. Here, however, the binomial used was Phytoxis acidissima, there being no mention of P. sideritifolia. It will be seen, then, that Molina (if he was also author of the appendix) applied two specific names to the same plant at different times.

Following the Kew rule, Bentham, in establishing his genus Sphacele, made the above-cited names synonymous with Sphacele campanulata, which binomial is still in general use. According to the usage of the International Code the second combination made by Briquet<sup>3</sup> is the proper one. Since the present case is illustrative of the manner in which an established and well-known name may be supplanted by one apparently never used save by its author, it is here suggested that the principle of usage embodied in the fifty-year rule of the International Code be extended to specific as well as generic names. Such application

<sup>&</sup>lt;sup>1</sup> Molina, I. I. Sagga sulla Storia Nat. del Chili, ed. 2, 145. 1810.

<sup>&</sup>lt;sup>2</sup> Feuillée, L. Hist. des Pl. Medecin., 4. 1725.

<sup>&</sup>lt;sup>3</sup> Briquet, J. in Engler und Prantl, Nat. Pflanzenfam., ed. 1, IV. Abt. 3 a, 291. 1897.

of this principle would certainly reduce in number undesirable but otherwise necessary changes in established and widely current nomenclature.

3. Sphacele subhastata Benth. in Edwards' Bot. Reg. 15: sub pl. 1289. 1829; Lab. Gen. et Sp. 569. 1834; et in DC. Prodr. 12: 255. 1848.

Alguelagum subhastatum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex ut videtur, ramis adscendentibus, superne floccosotomentosis, subteretibus, internodiis foliis subaequilongis; foliis 3-6 cm. longis, 8-14 mm. latis, oblongis, apice obtusis, in basi truncato-subhastatis, ad petiolum breviter cuneato-attenuatis, margine fere recta, crenata, crenarum culminibus circa 1 mm. altis, inter se 2-3 mm. distantibus, pagina superiore glabra, viride, rugosa sed non bullata, inferiore pallidiore, tomentosa, petiolis circa 1 cm. longis elatis; verticillastris 3-floribus, inter se 1-1.5 cm., bracteis internodiis paulo brevioribus, lanceolatis, acutis, subsessilibus, utrinque tomentellis; calycibus (?) florentibus 12-13 mm. longis, tubuloso-campanulatis, extus tomentellis, tubo turbinato, dentibus vix longiore, dentibus lanceolato-subulatis, subaequalibus, (?) maturis 18 mm. longis, forma fere immutatis, dentibus posticis ab anticis sinibus majoribus separatis; corollis nuculisque non visis.

Specimens examined:

Chili: in fruticetis calidis collium Quillota, Nov. 1829, Bertero 1017 (NY).

The above-listed specimen is the only one seen by the author which may clearly be referred to this species. The collection of Fielding (GH) resembles this in leaf outline, but is hardly otherwise separable from Sphacele Salviae; furthermore a portion of the same Bertero collection at the Missouri Botanical Garden appears to connect the two species. As observed by Bentham, the above-described plant is intermediate between S. Salviae and S. chamaedryoides and in foliage character and calyx suggests strongly the possibility of its being a hybrid between the two. According to Bentham the corolla is blue, 9–10 lines long, and broadly tubular-campanulate with stamens subexserted.

4. Sphacele Salviae Briq. in Bull. Lab. Bot. Gen. Genéve 1: 340. 1897.

Stachys Salviae Lindl. in Edwards' Bot. Reg. 15: sub pl. 1226. 1829.

Sphacele Lindlei [Lindleyi] Benth. in Edwards' Bot. Reg. 15: sub. pl. 1289. 1829.

Sphacele Lindleyi Benth. Lab. Gen. et Sp. 570. 1834; et in DC. Prodr. 12: 255. 1848.

Alguelagum Salviae Kuntze, Rev. Gen. 2: 511. 1891.

Frutex circa 1 m. altitudine, ramis erectis, teretibus, ramulis floccoso-lanatis, vix canaliculatis, angulis subacutis, internodiis quam foliorum longitudine paulo brevioribus; foliis 4-8 cm. longis, 2-4 cm. latis, lanceolatis, apice saepius obtusis, rarius breviter acuminatis, in basi truncato-cordatis subhastatisve, auriculis saepius rotundatis, margine revoluta, irregulariter crenato-dentata, crenarum culminibus circa 1-2 mm. altis. inter se 1.5-2.5 mm. distantibus, acutis, subapiculatis, pagina superiore viride bullato-rugosissima, dense sed minutissime tomentosa, inferiore albo-lanata, obscure reticulato-venulosa, petiolis lanatis, 1-2 cm. longis elatis; floribus in racemis simplicibus, rarius ramosis, albo-lanatis dispositis; verticillastris inter se 1-2 cm. distantibus, 1-3-floribus, oppositis, decussatim instructis, bracteis ovatis, subfoliosis, tomentosis, acutis vel breviter acuminatis, sessilibus, calycibus subaequilongis; calycibus tubulosis, floccoso-lanatis, florentibus 11-14 mm. longis. tubo 5-7 mm. longo, bilabiatis, labiis subaequalibus, dentibus labiarum superiorum 5 mm. longis, inferiorum 7 mm. longis, omnibus lanceolatis, setaceo-acuminatis, fructiferibus in basi leniter inflatis, 6-7 mm. latis, tubo 1 cm. longo; dentibus fere immutatis, erectis, sinibus subaequalibus separatis; corollis violaceis 2 cm. longis, tubulosis, fauce ampliatis, 5 mm. latis, ore obliquo, bilabiato, labiis subaequalibus, labiolo labro paulo longioribus, 4-5 mm. longo, lobis lateralibus medio dimidia parte brevioribus; staminibus didymis, ad faucem sitis, e tubo breviter exsertis, antheris 1.5 mm. longis, nectarostegio annulato e pilis densis 3 mm. supra tubi basim consistente: stylo breviter exserto: nuculis 2.5-3 mm. longis, atris, obovatis.

Specimens examined:

CHILI: ex collinis maritimis Chilensibus, prope Valparaiso, May 10, 1882, Ball (NY); Valparaiso, June 1885, Rusby 1399 (NY); Valparaiso, Feb. 1922, Bro. Claude-Joseph 1618 (US); Valdivia, 1862, Bridges (NY); in Gebüschen, Valparaiso, August 20, 1895, Buchtien (US); in fruticetis calidis collium Quillota, Nov. 1829, Bertero 1017 (MBG; GH; NY); near Valparaiso, 1851, Gillies (GH); near Valparaiso, Sept. 14, 1914, Rose 19115 (US; NY); Valparaiso, Wilkes Exped. (US; GH; ASP; NY); no data, Fielding (GH).

5. Sphacele speciosa St. Hil. in Benth. Lab. Gen. et Sp. 570. 1834; in DC. Prodr. 12: 254. 1848.

Alguelagum speciosum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex 1-1.5 m. altitudine, ramulis floccoso-tomentellis demum subglabris, obscure canaliculatis, angulis obtusis; foliis 5-7 cm. longis, 1.5-2.5 cm. latis, oblongo-lanceolatis apice obtusiusculis, junioribus in basi rotundatis, anguste truncato-subcordatis. maturis subsagittato-cordatis (Bentham), pagina superiore viride. glabra, bullata, inferiore dense et breviter rufo-lanata, reticulatovenulosa, pedicellis .5-1 cm. longis elatis; floribus in panicula laxa, ramulis gracilibus 5-10 cm. longis, in axillis bracteorum solitatim dispositis, decussatim instructis, nodis inter se 1-2 cm. distantibus, bracteis parvis, lanceolatis, acuminatis, deciduis subtentis; calveibus campanulatis, 13-venis, reticulato-venulosis, florentibus 12-13 mm. longis, tubo 6 mm. longo, venis hispidulis, dentibus aequilongis, lanceolato-acuminatis, bilabiatis, fructiferibus 18 mm. longis, tubo 10 mm. longo, plicato-venoso, ore 10-12 mm. lato, dentibus late patentibus, sinibus omnium subaequalibus; corollis 25 mm. longis, tubulosis, superne sensim ampliatis, bilabiatis, labiis aequilongis pro rata brevissimis, vix 2 mm., staminibus didymis, ad faucem sitis, filamentis anticis longis 6 mm., posticis dimidia parte brevioribus, antheris 1.5 mm. longis, nectarostegio antice supra tubi basim 4 mm. e pilis crassis consistente; stylo subexserto; nuculis 2.5 mm. longis, obovatis, atris.

Specimens examined:

Brazil: Serra da Itatiaia, retiro in campo May 15, 1902, Dusén 229 (US).

The type locality cited by Bentham is "Serra do Papagaio, Prov. Minas Geraes, in umbrosis rupestribus."

6. Sphacele lamiifolia Benth. Lab. Gen. et Sp. 570. 1834. Alquelagum lamiifolium Kuntze, Rev. Gen. 2: 511. 1891.

Frutex 1.5-2 m. altitudine, ramis teretibus, cortice discedente. ramulis rufo-lanatis, vix canaliculatis, quadratis, angulis subacutis, internodiis foliorum longitudine paulo brevioribus; foliis 8-11 cm. longis, 3-8 cm. latis, ovato-triangulis, apice obtusis sed tamen leniter acuminatis, in basi truncato-cordatis vel subsagittatis, margine irregulariter crenata, crenarum culminibus circa 1.5 mm. altis, inter se 3-6 mm. distantibus, pagina superiore viride. bullata, hispida, scabriuscula, inferiore subconcolore, subrufotomentosa vel sublanata, petiolis 1-1.5 cm. longis elatis; foliis supremis rotundato-ovatis, sessilibus; floribus in panicula laxa, ramulis gracilibus, rufo-villosis, angulis acutis dispositis; verticillastris 3-floribus, oppositis, decussatim instructis, distantibus, internodiis 1.5-2 cm. longis, bracteis rotundato-ovatis, tomentosis, apice breviter acuminatis, sessilibus, pedicellis quam calveibus brevioribus subtentis; calveibus campanulatis, scabriusculis, florentibus 12-14 mm. longis, purpurascentibus, fere Salviae, bilabiatis sed dentibus tamen aequilongis, 6 mm. longis, anguste lanceolatis, pungentibus, fructiferibus 2 cm. longis, late campanulatis, declinatis, chartaceis, ore obliquo, dentibus in basi 3-6 mm. latis, setaceo-acuminatis, rigidis, patentibus, vix recurvis, posticis tribus tamen ab anticis sinibus rotundatis latioribus separatis, pedicellis gracilibus 1 cm. longis elatis; corollis rubropurpurascentibus, 1.5-3 cm. longis, tubulosis, fauce .5-.7 mm. latis, superne gradatim dilatis, tubo recto vel leniter arcuato, ore obliquo, bilabiato, labiis subaequalibus, 2.5-3 mm. longis, labioli lobis lateralibus lobo medio brevioribus, staminibus didymis, 2.5-5 mm. longis, ad faucem sitis, antheris 1.5-1.7 mm. longis, nectarostegio annulato e pilis densis 1 mm. supra tubi basim consistente; stylo breviter exserto; nuculis atris, glabris, obovatis, 3 mm. longis.

Specimens examined:

PERU: no data, Weberbauer 5214 (FM); Wilkes Exped. (US; NY); Matucana, 2460 m., Apr. 12-May 3, 1922, Macbride & Featherstone 178 (MBG; FM).

7. Sphacele Hieronymi Briq. in Bull. de l'Herb. Boiss. 4: 806. 1896.

Suffrutex erectus, 60 cm. altitudine, caulibus villosulis, vix canaliculatis, angulis obtusis; foliis tenuibus, 4-7 cm. longis, 2.5-3.5 cm. latis, oblongis vel ovato-deltoideis, apice acutis, in basi truncato-hastatis, auriculis angularibus acutis vel subobtusis, margine longe et lenissime convexa, crenata, crenarum culminibus .5-1 mm. altis, inter se 2-4 mm. distantibus, pagina superiore viride, scabriuscula, inferiore praecipue ad venas tenuiter villosula, petiolis 1.5-2 cm. longis elatis; floribus in panicula laxa, puberula, ramulis adscendentibus dispositis; verticillastris 3-floribus, oppositis, decussatim instructis, inter se 1-2 mm. distantibus, bracteis .5-1 cm. longis, ovato-lanceolatis, acuminatis, pubescentibus, subintegris subtentis: calveibus campanulatis, pilis brevibus ad basim tenuiter vestitis, florentibus 7-8 mm. longis, tubo 4-5 mm. longo, dentibus 3-3.5 mm. longis, anguste lanceolato-acuminatis, fructiferibus 1.5 cm. longis, tubo 9 mm. longo, campanulato, reticulato-venuloso, dentibus 4-5 mm. longis, posticis tribus sinibus rotundatis latioribus ab anticis separatis, pedicellis 7 mm. longis elatis; corollis 9-14 mm. longis, late tubulosis, ad annulum constrictis, superne 4-5 mm. latis, bilabiatis, labiis lobisque subaequalibus circa 1 mm. longis, labioli lobo medio tamen lateralibus duplo latiore; staminibus ad tubi medium sitis, didymis, anticis 3-4 mm. longis, posticis dimidia parte brevioribus, antheris 1 mm. longis; nectarostegio annulato e pilis densis 2.5-3 mm., supra tubi basim sito, annulo infra sinum posticum interrupto; stylo breviter exserto; nuculis obovatis, 2.5 mm. longis, atris.

Specimens examined:

ARGENTINA: inter el Pan de Azucar et Colanchangua, Sierra Chica de Cordoba, Nov. 11, 1881, *Hieronymus 1005* (NY, type collection); Sierra de Chica, Cordoba, Dec. 6, 1876, *Hieronymus* (US); El Candado, Catamarca, Feb. 5, 1917, *Jörgensen 1264* (US).

This species is apparently conspecific with Sphacele floribunda Benth., of which no type locality is given, S. Grisebachii Kurtz<sup>2</sup>

<sup>&</sup>lt;sup>1</sup> Bentham, G. in DC. Prodr. 12: 254. 1848.

<sup>&</sup>lt;sup>2</sup> Kurtz, F. in Rev. de Museo de la Plata 5: 294. 1894.

(S. hastata Griseb. not Gray), Alguelagum Grisebachii Kuntze, and perhaps also with S. pampeana Speg.¹ According to Kurtz, S. Grisebachii occurs in two forms "una con el follage mas oscuro, tiene caliz y corolla mas o menos azulada; la otra, con sus hojas mas claras, posee flores de color rosado. La primera forma es la mas comun en nuestra Sierra, la segunda la he observado solamente una ves entre Copacabana y Avellaneda." The flowers of S. pampeana are white, according to Spegazzini, and while nearly related to S. hastata Griseb. the species may be readily distinguished from this "por los dientes del caliz mas largos del tubo del mismo."

## 8. Sphacele lancifolia (Rusby), comb. nov.

Alguelagum lancifolium Rusby in Bull. N. Y. Bot. Gard. 4: 434. 1907.

Frutex robustus, ramulis canaliculatis, subglabris, purpurascentibus, angulis obtusis; foliis 10-18 cm. longis, 3-5 cm. latis, lanceolatis vel elliptico-lanceolatis, apice acutis, rarius breviter acuminatis, in basi rotundatis et in petiolum 1-2 cm. longum extenuatis, margine crenata, crenarum culminibus circa 1 mm. altis, inter se 2.5-3 mm. distantibus, pagina superiore viride. glabra, subnitente, inferiore concolore, glabra, leniter reticulatovenulosa; floribus in panicula ramosiore saepe subternata approximatis; verticillastris 3-floribus, oppositis, densis, spirale instructis, bracteis lanceolatis, glabris, caerulescentibus, apice acuminatis, in basi subsessilibus, reticulato-venulosis, flores subaequantibus; calycibus late campanulatis, glabris, caerulescentibus. florentibus 9-12 mm. longis, tubo 5-6 mm. longo, 5-6 mm. latis, dentibus aequilongis, setaceo-acuminatis, pungentibus, pedicellis 2-3 mm. longis elatis, fructiferibus tubuloso-campanulatis tubo 8 mm. longo, dentibus 7-8 mm. longis, forma immutata; corollis 12-13 mm. longis, tubulosis sed superne patenter dilatis, fauce 5-6 mm. latis, bilabiatis, labiis lobisque tamen subaequalibus circa 2 mm. longis; staminibus didvmis, posticis 2-2.5 mm. longis, anticis duplo longioribus, omnibus ad tubi medium sitis, vix e tubo exsertis, antheris 1 mm. longis, nectarostegio subannulato e pilis densis supra tubi basim 3 mm. consistente,

<sup>&</sup>lt;sup>1</sup> Spegazzini, C. Flora Ventana, 49. 1896.

annulo infra sinum posticum interrupto; stylo vix exserto; nuculis atris, obovatis, 2 mm. longis.

Specimens examined:

Peru: San Miguel, Urubamba Valley, 1800 m., June 9, 1915, Cook & Gilbert 1144 (US).

BOLIVIA: no data, Bang 1823 (US; GH; ASP; MBG; NY, TYPE).

## 9. Sphacele hirsuta, sp. nov.

Suffrutex ramosus ut videtur, ramis ramulisque hirsutis, quadratis, angulis obtusis, internodiis folia saepius superantibus; foliis 2-5 cm. longis, 1.5-2 cm. latis, oblongis, obtusis, in basi rotundato-extenuatis in petiolum alatum longum .5-1.5 cm. productis, pagina superiore bullato-rugosa, viride, tenuiter pubescente, inferiore cinerea, hirsuta, venis tamen prominulis, margine convexiuscula, crenata, crenarum obtusarum culminibus 1 mm. altis, inter se 1-2 mm. distantibus; floribus in panicula hirsuta dispositis, verticillastris 3-floribus, oppositis, maturitate inter se 1.5-2 cm. distantibus, bracteis foliis conformibus sed minoribus subtentis; calveibus florentibus 3.5-4 mm. longis, dentibus subspinosis, fructiferibus 8-9 mm. longis, campanulatis, plicatovenosis, hirsutis, declinatis, dentibus 3 mm. longis, acuminatis, pungentibus, recurvis, tribus posticis sinibus rotundatis ab anticis separatis, pedicellis 3 mm. longis elatis; corollis maturis vix satis, ut videtur tamen calycibus aequilongis, staminibus parvis ad tubi medium sitis; nuculis circa 2 mm. longis, obovatis.

Specimens examined:

COLOMBIA: Párano near Bogotá, July 1917, Bro. Ariste-Joseph A 86 (US, TYPE).

Sphacele tenuifiora Benth. in DC. Prodr. 12: 257. 1848.
 Sphacele clinopodioides Griseb. in K. Ges. d. Wiss. Göttingen,
 Abh. 24: 273. 1879.

Alguelagum tenuiflorum Kuntze, Rev. Gen. 2: 511. 1891.

Suffrutex e rhizomate lignoso, 20-40 cm. altitudine, ramis ramulisque herbaceis, quadratis, canescenti-tomentosis, angulis obtusis, internodiis foliorum longitudine aequantibus vel paulo brevioribus; foliis 2-5 cm. longis, 1-4 cm. latis, saepius oblongis

interdum ovatis, apice obtusis, in basi rotundatis rarius subtruncatis, saepius in petiolum alatum brevem coarctatis, margine crenata, subrevoluta, crenarum culminibus .5-1 mm. altis, inter se 2-3 mm, distantibus, pagina superiore rugosa tenuiter canopubescente, inferiore pallide tomentosa, solummodo ad marginem reticulato-venulosa; floribus saepius in spicis vel glomerulis densis bracteosis confertis, spicis saepius pedunculatis, in fasciculis etiam corymbosis dispositis, foliis floralibus ovatis vel rotundatis, sessilibus, verticillastris oppositis, spirale instructis, bracteosis, 3-6-floribus, bracteis subfoliosis vel membranaceis, flores superantibus, ovatis vel lanceolato-oblongis apice saepe acutis, venulosis, tomentosis, margine integra vel interrupta; bracteolis membranaceis, oblongis vel linearibus; calycibus campanulato-tubulosis, pubescentibus, patenter glanduloso-punctatis, florentium tubo 2.5-3 mm. longo, 10-venis, ore obliquo, bilabiato, dentibus anguste lanceolatis, 1-1.5 mm. longis, fructiferum tubo 4-5 mm. longo, labiis subaequalibus 3-4 mm. longis, superiore declinata faucem claudente, dentibus tribus posticis sinibus acutis majoribus ab anticis separatis, omnibus ovato-triangularis, acutis, apiculatis, pedicellis 2 mm. longis; corollis albis, 6-8 mm. longis, tubulosis, superne paulo ampliatis, subglabris, bilabiatis, lobis subaequalibus, circa 1-1.5 mm. longis, labioli lobo medio 1.5-2 mm. longo, nectarostegio subannulato e pilis intus ad tubi basim consistente; staminibus tubo inclusis, supra medium positis; stylo tubum aequante; nuculis 1.7-2 mm. longis, atris, obovatis.

Specimens examined:

Bolivia: Unduavi, 2460 m., Oct. 1885, Rusby 1411 (NY); La Paz, Sonnige Abhänge, 3800 m., March, 1912, Buchtien 109 (GH; NY); Sorata, in graminosis, locis aridis, 2600-3100 m., March, 1860, Mandon 520 (GH; NY); no locality stated, 4200 m., Asplund 5879 (US); Yungas, 1890, Bang 167 (GH; US; ASP; MBG; FM; NY); Sorata, 3076 m., Feb. 1886, Rusby 1407 (MBG; US; ASP; GH); no data, Bang 1835 (GH; MBG; ASP; NY); near snow-line, Mt. Tunari, Cochabamba, 1891, Bang 1044 (US; MBG; ASP; FM; GH; NY); Lake Titicaca, 2840 m., Nov. 1910, Buchtien 2956 (US; NY); La Paz, 3800 m., May 6, 18, 1906, Buchtien, 131 (US; NY).

Peru: Ollantay-tambo, 3000 m., May 13, 1915, Cook & Gilbert 713 (US); Ollantay-tambo, 3000 m., July 18, 1915, Cook & Gilbert 1899 (US); Cuzco, 3000–3600 m., Herrara (US); Ollantay-tambo, 3000 m., May 18, 1918, Cook & Gilbert 801 (US).

ARGENTINA: Cuesta de Copina, Sierra Achala de Cordoba, Feb. 25, 1876, Hieronymus 441 (FM; US); Sierra de Tucuman, Jan. 10–17, 1874, Lorentz & Hieronymus 626 (US; FM; determined by Grisebach as S. clinopodioides Griseb.); Andalgalá, Cerra Negra, 3500 m., Feb. 19, 1916, Jörgensen 1307 (GH); same place and number, Feb. 20, 1917 (US); La Ciénega, Sierra de Tucuman, Jan. 10–17, 1874, Lorentz & Hieronymus 738 (NY, LOCOTYPE and probably type collection of Sphacele clinopodioides Griseb.).

11. Sphacele salviaefolia Benth. Lab. Gen. et Sp. 567. 1834; in DC. Prodr. 12: 256. 1848.

Sideritis salviaefolia Kunth in Humboldt & Bonpland, Nov. Gen. et Sp. Pl. 2: 307. 1817.

Alguelagum salviaefolium Kuntze, Rev. Gen. 2: 511. 1891.

Frutex ramis teretibus, cortice discedente: ramulis quadratis. canaliculatis, pulverulento-puberulis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 6-12 cm. longis, 2-5 cm. latis. lanceolato-oblongis, subacutis, in basi rotundato-truncatis, subinde ad petiolum coarctatis, margine crenata, subrevoluta, crenarum culminibus circa .1 mm. altis et inter se 2 mm. distantibus, regulariter dispersis, pagina superiore atro-viride, bullatorugosa, glabra, inferiore plumbea, pulchre reticulato-venulosa. minute et dense tomentosa, petiolis puberulis 1-1.5 cm. longis elatis; floribus in panicula saepius ternata, ramulis lateralibus 3-5 cm. longis in axillis foliorum summorum exstitis, ramo principale 5-8 cm. longo, 1.5-2 cm. lato, omnibus pedunculis 1-2 cm. longis elatis, villosulo-puberulis, verticillastris densis, oppositis, spirale instructis, 3-9 floribus, bracteis 5-10 mm. longis, submembranaceis, ovato-lanceolatis, acutis, reticulatovenulosis subtentis; calycibus tubulosis vel campanulato-tubulosis, villosulo-puberulis, his bracteisque saepius caerulescentibus, pedicellis 2 mm. longis elatis, fructiferibus 12 mm. longis, nutantibus, florentium tubo 4-5 mm. longo, 2.5-3.5 mm. lato, reticulatovenulosis, ore obliquo, vix bilabiato, dentibus 3–4 mm. longis, anguste lanceolato-acuminatis, acribus, subaequalibus; corollis albido-caerulescentibus, 7–9 mm. longis, 2–3 mm. latis, campanulato-tubulosis, villosulo-puberulis, in facie inferiore prope annulum paulo sacculatis, sub-bilabiatis, labro bifido, lobis 1 mm. longis, labioli lobo medio paulo longiore; nectarostegio annulato imperfecto e pilis consistente supra tubi basim 2–3 mm. posito; staminibus minutissimis, 1–1.5 mm. longis, subaequalibus, in tubo inclusis, prope medium sitis, antheris 1.3 mm. longis; stylo tubum subaequante; nuculis siccis 1.7 mm. longis, 1.3 mm. latis, obovato-oblongis, atris, hebetibus, apice rotundatis, basi subtruncatis, hilo obscuro.

Specimens examined:

COLOMBIA: Bogotá, 1918, Bro. Ariste-Joseph A 306 (US); bushy slope, base of mountain, 2800–2900 m., Oct. 4–8, 1917, Pennell 2376 (US; MBG; FM; GH); near Bogotá, 2800 m., Idinael 10 (NY); Bogotá, 1919, Bro. Ariste-Joseph (US); Sabana de Bogotá, May 1923, Pring 102 (MBG); in montibus juxta Bogotam, Nov. 10, 1852, Holton 486 (GH; NY).

12. Sphacele inflata Brig. in Bull. de l'Herb. Boiss. 4:848. 1896. Frutex ramulis quadratis, canaliculatis, puberulis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 8-10 cm. longis, 2-3.5 cm. latis, tenuibus, anguste oblongo-lanceolatis, leniter acuminatis, in basi rotundato-coarctatis, margine crenatoserrata, crenarum culminibus circa .5 mm. altis, et inter se 1.5-2 mm. distantibus, regulariter dispersis, pagina superiore viride leniter bullata, glabra, inferiore rufo-tomentosa, obscure reticulato-venulosa, petiolis tomentosis .5-1.0 cm. longis elatis; floribus in panicula ramulis inferioribus adscendentibus, 5 cm. longis, omnibus rufo-tomentosis; verticillastris densis, infra approximatis, oppositis, spirale instructis, 6-floribus, bracteis 3-5 mm. longis, ovato-lanceolatis, acuminatis, submembranaceis, tomentosis; calycibus florentibus 3-4 mm. longis, campanulatotubulosis, pedicellatis, tomentosis, tubo 1.5-2 mm. longo, dentibus subaequalibus, 1-1.5 mm. longis, fructiferum tubo circa 6 mm. longo, chartacec-inflato, purpurascentibus, dentibus erectis, acuminatis, 1-1.5 mm. longis; corollis longis 3.5-4 mm., tubulosis,

superne sensim ampliatis, glabris, tubo 2.5-3 mm. longo, nectarostegio inannulato e pilis areolam inter staminum bases facientibus consistente, limbo subbilabiato lobis circa 1 mm. longis, subaequalibus, labioli lobo medio 1.5 mm. longo; staminibus didymis, anticis subexsertis, ad tubi medium sitis, stylo corollam subaequante; nuculis ovato-oblongis, fuscis,  $1.3 \times .8$  mm.

Specimens examined:

BOLIVIA: no locality stated, 2600 m., Apr. 13–21, 1892, *Kuntze* (NY, ? TYPE); Sorata, 2460 m., Feb. 1886, *Rusby 1416* (US; MBG; ASP; GH; NY).

The above description is based upon a specimen in the Kuntze Herbarium at the New York Botanical Garden which corresponds in every particular to Briquet's description. Briquet in that instance failed to cite the specimen before him. It is probable, however, that this is the type. If so, it appears to be an imperfect or immature specimen of the species illustrated by the beautiful series of the Rusby collection. This plant accords well with the Kuntze plant but the foliage attains a greater size, the panicle is much larger, being much branched and reaching a length of twenty centimeters and an equal width at the base. The calyx, furthermore, is larger in fruit, the tube being 7–8 mm. long and nearly as wide, the teeth being 2.5–3 mm. long. The mature calyx resembles that of S. aurifera but is not as large and the teeth are notably narrower and acuminate.

## 13. Sphacele aurifera (Rusby), comb. nov.

Alguelagum auriferum Rusby, Mem. Torrey Bot. Club 5: 108. 1891.

Sphacele Kuntzeana Briq. in Bull. de l'Herb. Boiss. 4: 805. 1896.

Sphacele cochabambana Briq. loc. cit. 807. 1896.

Sphacele confusa Briq. loc. cit. 806. 1896

Frutex robustus ramulis quadratis, subcano-tomentosis, canaliculatis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 10-15 cm. longis, 3.5-5 cm. latis, lanceolatis, apice acutis vel longe leniterque acuminatis, in basi rotundatis, margine crenato-serrata, crenis acutis apiculatis, culminibus circa 1.5 mm. altis et inter se 2-3 mm. distantibus, pagina superiore viride,

glabra, bullato-rugosa, inferiore pallidiore rufula, reticulatovenulosa, sparse tomentella, glandulis aureis frequenter prominentioribus, petiolis 1-1.5 cm. longis elatis; floribus in panicula ampla, ramulis cano-tomentosis, adscendentibus dispositis; verticillastris densis, fructiferibus confertis, 6-floribus, oppositis, spirale instructis; bracteis membranaceis saepe caerulescentibus flores subaequantibus vel superantibus, acuminatis, reticulatovenulosis; calycibus membranaceis, glabris, ore obliquo, vix bilabiato, florentibus tubuloso-campanulatis, tubo 3-4 mm. longo, dentibus 1-1.5 mm. longis, acuminatis, fructiferibus inflato-campanulatis, 12-14 mm. longis, tenuibus, patente reticulatovenulosis, dentibus ovato-triangulis, acutis, subapiculatis, 3.5-4 mm. longis, corollis 4-5 mm. longis, tubulosis, bilabiatis, lobis subaequalibus, labioli lobo medio tamen longiore, tubo 3.5-4 mm. longo, vix annulato sed e medio ad faucem pilis longis ornatis, staminibus .5 mm. longis, inclusis, supra medium sitis; stylo subexserto; nuculis 2 mm. longis, obovatis, atris.

Specimens examined:

Bolivia: near snow-line, Mt. Tunari, Cochabamba, 1891, Bang 1107 (US; MBG; ASP; GH; NY, TYPE); Cochabamba, 3000 m., March 26, 1892, Kuntze (NY, type collection of Sphacele cochabambana Briq.); Cochabamba, 3000 m., March 26, 1892, Kuntze (NY, type collection of Sphacele Kuntzeana Briq.); near snow-line, Mt. Tunari, Cochabamba, 1891, Bang 1108 (US; MBG; ASP; GH; NY); no locality stated, 3800 m., March 18, 1892, Kuntze (NY, type collection of Sphacele confusa Briq.).

All specimens above cited are apparently from the same locality and while varying considerably in superficial aspect can hardly be considered distinct. S. Kuntzeana and S. aurifera are both based on specimens in full fruit and are unquestionably the same. S. cochabambana is a specimen just coming into flower in which the leaves are almost as broad as long. Only the uppermost internodes are present. As suggested by Briquet it may prove to be a variety. S. confusa appears to the author to be based on nothing other than a flowering specimen of S. aurifera in which no calyces have become mature. By reason of the large size of the mature calyx and the resultant crowding, a fully fruiting branch has an appearance quite different from a

branch in flower. The relationship of this species with Sphacele inflata Briq. is close but uncertain to the author.

14. Sphacele parviflora Benth. in DC. Prodr. 12: 256. 1848. Alguelagum parviflorum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex circa 1 m. altitudine, ramis teretibus, cortice discedente: ramulis quadratis, canaliculatis, tomentoso-puberulis, subrufis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 10-18 cm. longis, 4-6 cm. latis, lanceolato-ellipticis, breviter acuminatis, in basi ad petiolum attenuatis, margine crenata, subrevoluta, crenarum culminibus circa 1 mm. altis et inter se 2 mm. distantibus, regulariter dispersis, pagina superiore atroviride bullato-rugosa, glabra, inferiore subrufa, reticulato-venulosa, minute et dense tomentosa, petiolis puberulis, 1-1.5 cm. longis elatis; floribus in panicula ramulosa 12-20 cm. longa exstita, ramulis inferioribus adscendentibus, 8-12 cm. longis, omnibus rufo-pubescentibus; verticillastris densis infra approximatis, oppositis, spirale instructis, 6-floribus, bracteis flores subaequantibus, ovatis vel lanceolatis, subacuminatis, in basi saepius coarctatis, submembranaceis, reticulato-venulosis sed vena media venisque parallelis tamen prominentibus; calycibus florentibus campanulato-tubulosis, tubo 3-4 mm. longo, reticulato-venulosis, villosulo-puberulis, ore obliquo, subbilabiato, dentibus subaequalibus, tribus posticis in basi connatis. lanceolato-acuminatis, 1 mm. longis, fructiferum tubo 6-7 mm. longo, chartaceo-inflato, fauce paulo constricto, dentibus 1.5-2 mm., subconniventibus; corollis albis, 4-5 mm. longis, tubulosis, supra sensim ampliatis, villosulo-puberulis, tubo 3-3.5 mm. longo, vix annulato, nectarostegio tamen e pilis ad bases staminum inferiorum areolam facientibus consistente, ore obliquo, subbilabiato, lobis 1.5 mm. longis, subaequalibus, labioli lobo medio 2 mm. longo, patulo; staminibus minutis, 1.5-2 mm. longis, subaequalibus, prope tubi medium positis; antheris .5 mm. longis, e tubo vix exsertis; stylo tubum subaequante; nuculis oboyato-oblongis, atris,  $1.3 \times .8$  mm.

Specimens examined:

COLOMBIA: Popayan, Lehmann 806 (NY); no data, Lehmann 5504 (US); hillside field, 1800-2100 m., Salento, Caldas, July

25-31, 1922, Pennell, Killip & Hazen 8747 (ASP; NY); thicket growth (machimbi), 1700-1900 m., Cuatro Esquinas to Rio Piendamo, El Cauca, June 6, 1922, Pennell & Killip 6398 (US; ASP); thicket growth, 1700-1900 m., Cuatro Esquinas to Rio Piendamo, El Cauca, June 6, 1922, Pennell & Killip 6387 (ASP); bushy banks, 2200-2400 m., Salento, June 27, 1922, Pennell & Killip 7278 (US; ASP; NY); thicket growth, 1700-1900 m., Cuatro Esquinas to Rio Piendamo, El Cauca, June 6, 1922, Pennell & Killip 6383 (US; ASP; NY).

VENEZUELA: Paramos between St. Domingo and Chacopo, Merida, 3300 m., Jahn 1129 (US); Silla de Caracas, 2460 m., May 21, 1874, Kuntze Herb. 1658 (NY); prope coloniam Tovar, 1854-5, Fendler 868 (MBG; NY); Agua de Obispo, 2500 m., Sept. 24, 1922, Jahn 1170 (US).

- S. Lindeniana Briq. may be referable here. Briquet's description was based upon a specimen in fruit with no flowers. From the description it seems hardly distinct from the forms of S. parviflora which grow in this region, which are quite variable in foliage character, particularly with reference to size.
- 15. Sphacele acuminata Grisebach in K. Ges. d. Wiss. Göttingen, Abh. 19: 238. 1874.

Alguelagum acuminatum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex vel arbuscula aromatica foliosa ramosiaque 2-4 m. altitudine, ramulis quadratis, canaliculatis, angulis obtusis; foliis 9-15 cm. longis, 2.5-5.5 cm. latis, membranaceis, lanceolatis, apice leniter acuminatis, in basi ad petiolum coarctatis, vix attenuatis, margine regulariter serrato-crenata, crenarum culminibus 1-1.5 mm. altis, pagina superiore subscabra, vix bullata, subglabra, inferiore pallida, tomentella, petiolis tomentosis 1-1.5 cm. longis elatis; floribus in panicula diffusiore, 15 cm. longa, 15-20 cm. lata, pyramidata, ramulis floccoso-tomentosis, lateralibus valde divaricatis dispositis; verticillastris 2-6-floribus, oppositis, spirale instructis, inter se .5 cm. distantibus; bracteis membranaceis, quam floribus brevioribus, ovatis, tomentosis, reticulato-venulosis subtentis; calycibus turbinato-campanulatis, membranaceis, puberulis, pedicellis 1.5 mm. longis elatis, florentium tubo 1.5 mm. longo, dentibus 1 mm. longis, lanceolato-

acuminatis, fructiferibus subcampanulatis, tubo 5-6 mm. longo, ore obliquo, subbilabiato, dentibus 2-3 mm. longis, triangulo-ovatis, acutis; corollis albis, tubulosis superne vix ampliatis, 4 mm. longis, lobis subaequalibus, .5-7 mm. longis, staminibus minutissimis, .4 mm. longis, supra tubi medium insertis, antheris filamentis aequilongis, nectarostegio e pilis ad bases filamentorum areolas duas facientibus consistente; stylo e corolla breviter exserto; nuculis non visis.

Specimens examined:

ARGENTINA: Andalgalá, Feb. 1915, Jörgensen 1265 (MBG); same locality and number, Sept. 2, 1915 (GH); same locality and number, Apr. 2, 1917 (US).

# 16. Sphacele Mandoniana Briq. Ann. Conserv. Genéve 2: 176. 1898.

Frutex ramulis quadratis, canaliculatis, puberulis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 10-15 cm. longis, 2-4 cm. latis, anguste lanceolatis, leniter acuminatis in basi ad petiolum attenuatis, margine crenato-serrata, subrevoluta, crenarum culminibus circa 1 mm. altis et inter se 1.5-2 mm. distantibus, regulariter dispersis, pagina superiore viride, leniter bullata, glabra, inferiore puberula, obscure reticulatovenulosa, petiolis puberulis, .7-1.0 cm. longis elatis; floribus in panicula ramosiore 12-20 cm. longa, ramulis inferioribus divergentibus 10-12 cm. longis, omnibus puberulis; verticillastris oppositis, spirale instructis, inter se circa .5 cm. distantibus, infra 5-floribus, supra 3-floribus, bracteis 2-4 mm. longis, ovatis, acuminatis, puberulis, submembranaceis; calycibus florentibus 3 mm. longis, campanulatis, tubo 1.5-2 mm. longo, reticulatovenulosis, puberulis, dentibus subaequalibus, 1-1.5 mm. longis, subulatis, patulis, fructiferum tubo 6-7 mm. longo, chartaceoampliato, fauce haud constricto, dentibus 1.5-2 mm. longis; corollis 4-5 mm. longis, tubulosis, superne sensim ampliatis, glabris. tubo 3-3.5 mm. longo, nectarostegio e pilis ad staminum inferiorum bases areolam facientibus consistente, subbilabiatis, lobis 1-1.5 mm. longis, subaequalibus, labioli lobo medio 1.5 mm. longo; staminibus minutis, 1.5 mm. longis, subaequalibus, ad tubi medium positis; antheris .5 mm. longis, e tubo vix exsertis,

stylo tubum subaequante; nuculis obovato-oblongis, fuscis, 1.3 × 8 mm.

Specimens examined:

Bolivia: Prov. Larecaja, viciniis Sorata, Espada in dumosis, 2600 m., March, 1889, Mandon 505 (GH, type collection); Yungas, 1890, Bang 686 (US; MBG; ASP; GH; NY); Unduavi, Nord Yungas, 3300 m., Nov. 1910, Buchtien 317 (US).

# 17. Sphacele heteromorpha Briq. in Bull. de l'Herb. Boiss. 4: 847. 1896.

Frutex robustus, vel arbuscula 2 m. et ultra altitudine, ramulis rufo-tomentosis, canaliculatis, internodiis minus quam foliorum latitudine, angulis obtusis; foliis maximam partem magnis, 10-18 cm. longis, 5-8 cm. latis, submembranaceis, saepius ovatoellipticis, apice acutis vel breviter acuminatis, in basi saepius rotundato-truncatis rarius in petiolum extenuatis vel subcordatis. margine irregulariter crenata, crenarum culminibus circa .5-1 mm. altis, inter se 2-3 mm. distantibus, pagina superiore viride, glabra, bullato-rugosa, inferiore tenuiter rufo-tomentellis, patenter reticulato-venulosis, petiolis 2-4 cm. longis elatis, foliis juvenalibus elongato-oblongis, subtus valde rufo-pannosis; floribus in panicula ramulis rufo-tomentosis subfasciatis dispositis, verticillastris densis vix interruptis, oppositis, subdecussatim instructis, bracteis ovatis, calycibus aequilongis vel subduplo longioribus, acutis, sessilibus, tomentosis, reticulato-venulosis sed venis parallelis tamen prominentibus, bracteolis parvis, linearibus, plerisque duobus: calvcibus tubuloso-campanulatis, tomentellis, florentium tubo 2.5 mm. longo, subsessilibus, intus ad basim pubescente, ore obliquo, bilabiato, dentibus lanceolatis, acutis, subapiculatis, 1-1.3 mm. longis, tribus posticis in basi connatis, maturis 5-6 mm. longis, tubulosis, tubo 4-4.5 mm. longo, dentibus fere immutatis; corollis 4-7 mm. longis, tubulosis, superne ampliatis, bilabiatis, lobiis subaequalibus, labro profunde bifido, lobis rotundatis, lobis labioli lateralibus lobo medio duplo brevioribus; staminibus parvis 1-2.5 mm. longis, ad tubi medium sitis, nectarostegio annulato e pilis densis ad bases filamentorum praecipue anticorum consistente; stylo corollam aequante: nuculis non visis.

Specimens examined:

Bolivia: no locality stated, 1600 m., Apr. 13-21, 1892, Kuntze (NY, type collection); Unduavi, Nordyungas, 3200 m., Feb. 1914, Buchtien (GH); Yungas, 1890, Bang 689 (ASP; US; MBG; GH; NY, Alguelagum confertum Rusby non Kuntze); Unduavi, 2460 m., Oct. 1885, Rusby 1415 (ASP; GH; US; NY, Alguelagum confertum Rusby non Kuntze).

18. Sphacele conferta Benth. Plantae Hartweg. 244. 1846; et in DC. Prodr. 12: 256. 1848.

Alguelagum confertum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex circa 1 m. altitudine, ramulis quadratis, puberulis, canaliculatis, angulis obtusis, foliis 8-12 cm. longis, 4-5.3 cm. latis, oblongo-lanceolatis, apice acutis, in basi truncato-subsagittatis, margine revoluta crenato-dentata, longe leniterque convexa, crenarum culminibus acutis, circa 1-1.5 mm. altis, inter se 1.5-2 cm. distantibus, pagina superiore atro-viride, glabra. bullata, inferiore reticulato-venulosa, cano-tomentosa, petiolis puberulis 1.5-2 cm. longis elatis; floribus in panicula, ramulis rufo-tomentosis, adscendentibus, 4-6 cm. longis, congestis; verticillastris confertis, oppositis, spirale instructis, 6-floribus, bracteis calyces maturos aequantibus vel superantibus, ovatis, obtusis, puberulis, sessilibus subtentis; calycibus tubulosis, puberulis, 10-venis, membranaceis, ore obliquo, breviter bilabiato, florentium tubo 2.5-3 mm. longo, dentibus circa 1 mm. longis, acutis obtusiusculisque, fructiferibus 7 mm. longis, dentibus 1.5-2 mm. longis, ovato-triangulis, acutis vel obtusis, tribus posticis longioribus, pedicellis 1 mm. longis elatis; corollis 6 mm. longis, bilabiatis, lobis subaequalibus, labioli lobo medio tamen lateralibus duplo longiore, tubo 4 mm. longo, sensim superne dilatis, nectarostegio e pilis ad filamentorum anticorum bases consistente; staminibus posticis 1.5 mm. longis, anticis majoribus, 2 mm. longis, omnibus ad tubi medium sitis, stylo subexsertis; nuculis 1.5 mm. longis, oblongo-obovatis, atris.

Specimens examined:

COLOMBIA: prope pagum Pitayo, Prov. Popayan, Hartweg 1348 (NY, type collection); San Cristobal, Nov. 1911, Bro. Apollinaire & Bro. Arthur 62 (US); open near Rio Anambiu, El Cauca, 2900-3200 m., June 11-16, 1922, Killip 6763 (ASP; US).

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PERU: sunny canyons, 2460 m., 7 mi. s. w. of Panao, May 8, 1923, Macbride 3575 (US; MBG).

The above-cited specimen collected by Macbride is referred here with some hesitation. The Hartweg collection at the New York Botanical Garden, upon which the above description is largely based, has decidedly truncate leaves which are mostly subsagittate, the auricles when present being about 5 mm. long, divaricate and acute, the blade being somewhat narrowed above them, expanding toward the middle. The leaves of the Killip collection are similar but less pronounced in the sagittate character and slightly cordate. The leaves of the collection made by Bro. Apollinaire and Bro. Arthur are only occasionally subsagittate but are mostly cordate. All are of about the same texture. The foliage of the Peruvian plant collected by Macbride is very similar to the last-mentioned Colombian plant, but is more densely cano-tomentose on the lower surface. While the calyces of the Colombian plants are usually obtuse, they are distinctly acute on the Killip plant, which is otherwise hardly separable from the type. In the Macbride collection, however, the calvx teeth are distinctly acute and even somewhat acuminate, the other flower characters being about the same as those of the Colombian plants. The species is apparently a variable one and may very probably include S. bullata Kunth and S. paniculata Kunth, the descriptions of which suggest similar plants. The author has been unable to identify definitely any collections seen by him with either of these two species, however. S. intermedia, a species herein newly described is closely allied and may prove conspecific. The calyces of this, however, are nearly those of S. mutica.

## 19. Sphacele intermedia, sp. nov.

Frutex altitudine circa 2 m., ramis teretibus, cortice discedente, ramulis floccoso-tomentosis, canescentibus, quadratis, canaliculatis, angulis obtusis; foliis 4-8 cm. longis, 2-3 cm. latis, oblongis, apice obtusis, in basi truncato-subcordatis, pagina superiore subcinerea (in spec. siccis), subglabra, ad venas tomentella, rugoso-bullata, inferiore pallidiore, canescenti-tomentosa, obscure reticulato-venulosa, margine convexiuscula, subrevoluta,

crenata, crenarum culminibus .5-1 mm. altis, inter se 1.5-2 mm. distantibus, petiolis floccoso-tomentosis 1 cm. longis elatis; floribus in panicula, ramulis adscendentibus, canescentibus, 3-5 cm. longis congestis; verticillastris confertis, sub-6-floribus, vix oppositis, spirale instructis, bracteis rotundatis, obtusis, infra tomentosis, supra glabris, sessilibus subtentis; calycibus subsessilibus, membranaceis, canescenti-tomentosis, ore obliquo, patenter bilabiato, labiae superioris lobis brevissimis obtusis. florentium tubo 2.5 mm. longo, dentibus inferioribus, ovatotriangulis, 1.5 mm. longis, labia superiore paulo breviore, fructiferibus pallide violaceis, tubo 4 mm. longo, dentibus fere immutatis; corollis (?) albis. 3.5-4 mm. longis, calucem vix superantibus. bilabiatis, lobis subaequalibus, labioli medio lateralibus tamen duplo longiore, tubo 3 mm. longo, nectarostegio e pilis ad filamentorum anticorum bases areolam facientibus consistente; staminibus .8 mm. longis, ad tubi medium sitis, antheris pro floris magnitudine magnis; stylo vix exserto; nuculis non visis.

Specimens examined:

Ecuador: Otavalo to Malchingui, 2400–3000 m., April 12, 1923, *Hitchcock 20839* (US, TYPE); vicinity of Tablón de Ona, Sept. 27, 1918, *Rose 23068* (NY).

A plant closely allied to both S. conferta and S. mutica. In the shape of the leaves it suggests S. Sprucei; in texture and pubescence, however, its foliage resembles that of S. conferta; in flower character and habit it is close to S. mutica.

# 20. Sphacele Sprucei Briq. Ann. Conserv. Genéve 2: 178. 1898.

Frutex ut videtur elatus, ramulis undique dense floccoso-tomentosis, canaliculatis, quadratis, angulis obtusis; foliis oblongis, obtusis, in basi truncato-constrictis, in petiolum brevem decurrentibus, margine longe leniterque convexiuscula, revoluta, crenata, culminibus .5 mm. altis, inter se 2 mm. distantibus, obtusis, subapiculatis, pagina superiore glabra, viride, bullata, inferiore crispulo-tomentella, venis evidentioribus, his petiolisque 1-1.5 cm. longis, floccoso-tomentosis; floribus in panicula, ramulis divergentibus, floccoso-tomentellis; verticillastris sub-6-floribus, densis, globosis, oppositis, moniliformis, inter se .5-1 cm. distanti-

bus; bracteis rotundatis, tomentosis, calycibus aequilongis; calycibus florentibus 3 mm. longis, subsessilibus, turbinato-campanulatis, tomentosis, bilabiatis, dentibus posticis in basi connatis, omnibus tamen obtusis, aequilongis, fructiferibus 4–5 mm. longis, campanulatis, dentibus majoribus sed forma immutatis; corollis 3.5 mm. longis, e calyce vix exsertis, tubo 2.5–3.5 mm. longo, nectarostegio e pilis inter filamentorum anticorum bases areolam facientibus consistente, labro bifido, lobis eis labioli lateralibus aequilongis, lobo medio duplo longiore; staminibus didymis, paulo supra tubi medium insertis, antheris e tubo exsertis; stylo in tubo incluso; nuculis obovatis, atris, 1.3 mm. longis.

Specimens examined:

Ecuador: in Andibus, 1857-9, Spruce 6090 (GH, type collection).

21. Sphacele cordifolia Benth. in DC. Prodr. 12: 257. 1848. Alguelagum cordifolium Kuntze, Rev. Gen. 2: 511. 1891.

Frutex ramulis rufo-lanatis, canaliculatis, quadratis, angulis obtusis, internodiis latitudinem foliorum subaequantibus: foliis 5-9 cm. longis, 3-5 cm. latis, ovato-oblongis, obtusis, tamen leniter acuminatis, basi patenter cordatis, lobis circa 1 cm. longis, rotundatis, margine crenata, crenarum culminibus .5-1 mm. altis, inter se 2-2.5 mm. distantibus, pagina superiore atro-viride. bullato-rugosa, hirtella, pagina inferiore pallidiore, lanato-tomentosa, petiolis 1.5-2 cm. longis, lanato-pubescentibus elatis; floribus in panicula ramulis rufo-lanatis, divergentibus, 3-6 cm. longis, infimis iterum ramosis dispositis; verticillastris densis vix interruptis, sub-6-floribus, oppositis, spirale instructis, bracteis ovato-rotundatis, tomentosis, sessilibus, flores subaequantibus. venis parallelis, apice saepe breviter bidentatis subtentis; calycibus campanulato-tubulosis, hirtellis, subsessilibus, florentium tubo longo 3 mm., ore obliquo patenter bilabiato, dentibus aequilongis, sed posticis in basi connatis, omnibus lanceolatis, acutis, subapiculatis, patulis, 1.5-2 mm. longis; corollis 5-6 mm. longis, tubulosis, superne paulo dilatis, tubo 4-4.5 mm. longo, ore bilabiato, labiolo labro triplo longiore, lobo medio patulo, 1.7 mm. longo, staminibus didymis, posticis 1 mm. longis, supra

tubi medium sitis, anticis 2 mm. longis, ad tubi medium sitis, antheris subaequalibus, e tubo breviter exsertis, nectarostegio e pilis ad filamentorum anticorum bases consistente; stylo tubum subaequante; nuculis non visis.

Specimens examined:

Peru: above Ayavaca, Piura, 2900-3000 m., May 1912, Weberbauer 6368 (FM).

22. Sphacele radula Benth. in DC. Prodr. 12: 257. 1848. Alguelagum radulum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex 2 m. altitudine, ramulis quadratis, canaliculatis. floccoso-tomentosis, sublanatis, angulis obtusis; foliis 7-15 cm. longis, 4-7 cm. latis, ovato-ellipticis apice obtusis, in basi rotundatis, truncato-subcordatis, pagina superiore atro-viride bullato-rugosa. glabra, inferiore praecipue in foliis juvenalibus rufo-tomentosa. sublanata, margine crenato-serrata, crenarum culminibus 1-1.5 mm. altis, inter se 2-3 mm. distantibus, petiolis 2 cm. longis in basi patenter dilatis, rufo-tomentosis elatis; floribus in panicula ramulis rufo-tomentosis dispositis; verticillastris densis, vix interruptis, 6-9-floribus, oppositis, subdecussatim instructis, bracteis ovatis, sessilibus, obtusis, membranaceis, tomentosis, reticulatovenulosis, vena media solummodo prominentiore subtentis; calveibus tubuloso-campanulatis, tomentellis, florentibus 4 mm. longis, subsessilibus, ore oblique, bilabiato, labia inferiore breviore, dentibus subaequalibus 1 mm. longis, obtusis; corollis 4.5-5 mm. longis, tubo superne patenter ampliato, bilabiatis, lobis subaequilongis, labioli lobo medio tamen lateralibus duplo longiore, staminibus subdidymis ad tubi medium sitis, saepe 3 mm. longis, nectarostegio e pilis ad filamentorum anticorum bases areolam facientibus consistente; stylo corollam aequante; nuculis non visis.

Specimens examined:

Ecuador: Loja, between San Lucas and Ona, 2200-3100 m., Sept. 7, 1923, *Hitchcock 21577* (US).

23. Sphacele mutica Benth. Pl. Hartweg. 145. 1844; in DC. Prodr. 12: 256. 1848.

Alguelagum muticum Kuntze, Rev. Gen. 2: 511. 1891.

Frutex 1-2 m. altitudine, ramulis quadratis, obscure canaliculatis, canescenti-puberulis, angulis obtusis; foliis 5-9 cm. longis, 1-2 cm. latis, oblongo-lanceolatis, apice obtusis vel acutiusculis, in basi ad petiolum coarctatis, margine revoluta. obscure crenata, crenarum culminibus .2-.3 mm. altis, inter se 1.5-2 mm. distantibus, pagina superiore rugoso-bullata, bullis parvis, molliter puberula, inferiore plumbea, pulchre reticulatovenulosa, minute tomentosa praecipue et dense in foliis juvenalibus, petiolis longis .5 cm. elatis; floribus in panicula 6-12 cm. longa ramulis 3-8 cm. longis, tomentosis dispositis; verticillastris sub-6-floribus, oppositis, dense et spirale instructis, bracteis parvis ovato-rotundatis, tomentosis, sessilibus, flores vix aequantibus subtentis; calycibus tubuloso-campanulatis sub-5-venis canescenti-tomentosis, florentibus 3 mm. longis, dentibus .5-1 mm. longis, obtusis, fructiferibus 5 mm. longis, subinflatis vix bilabiatis, dentibus immutatis, pedicellis 1 mm. longis elatis: corollis tubulosis, superne paulo ampliatis, 3.5-4 mm. longis, lobis parvissimis, subaequalibus, labioli lobo medio tamen lateralibus duplo longiore, .7 mm. longo; staminibus parvis, inclusis, ad tubi medium sitis, nectarostegio e pilis ad filamentorum anticorum bases areolam facientibus consistente; stylo in tubo incluso; nuculis (?) maturis obovato-oblongis, fuscis, 1.5 mm. longis.

Specimens examined:

Ecuador: in montibus, Loxa (Loja), Hartweg 809 (NY, type collection); no data, Jameson (US); Loja, Sept. 29-Oct. 3, 1918, Rose 23277 (NY); Loja, no date, Lehmann 4954 (NY).

# 24. Sphacele mollis sp. nov.

Frutex ramis teretibus, cortice discedente, ramulis canaliculatis, minute tomentosis, quadratis, angulis obtusis, internodiis minus quam foliorum longitudine; foliis 3–8 cm. longis, 1.5–5 cm. latis, oblongis, utrinque acutis subaequaliter coarctatis, margine obscure crenata vel subintegra, pagina superiore atro-viride (in spec. siccis) minutissime puberulis, inferiore pallidiore, tomento densiore vestita, petiolis puberulis .5 cm. longis elatis; floribus in panicula diffusiore dispositis, ramulis gracilibus, puberulis, divaricatis, 4–6 cm. longis, verticillastris oppositis, spirale in-

structis, densioribus, 6-9 floribus, inter se .5-1 cm. distantibus, bracteis flores paulo superantibus, ovatis, acutis, in basi angustatis, foliosis, reticulato-venulosis, puberulis; calycibus campanulatis, praesertim ad venis puberulis, florentibus 2.5 mm. longis, tubo 2 mm. longo, ore obliquo, subbilabiato, dentibus tamen subaequalibus, acutis, minute apiculatis, (?) maturis 5 mm. longis, paulo inflatis, ore vix constricto, dentibus 1.5 mm. longis, pedicellis 2 mm. longis elatis, corollis tubulosis superne leniter ampliatis, bilabiatis, labiis lobisque subaequalibus labioli lobo medio tamen longioribus, tubo 2 mm. longis, e calyce vix exserto, staminibus minutissimis, .3 mm. longis, ad tubi medium sitis, nectarostegio e pilis longis sparsis intra filamentorum anticorum bases areolam facientibus consistente; stylo tubum subaequante; nuculis (?) maturis, atris, obovatis, 1.2 mm. longis.

Specimens examined:

Peru: above Olmos, 1900-2000 m., Prov. Lambeyeque, May 1915, Weberbauer 7106 (FM, TYPE).

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# EXPLANATION OF PLATE

PLATE 6

Sphacele hirsuta Epling

Colombia

From the type specimen, Bro. Ariste-Joseph A 86, in the United States National Herbarium.



EPLING SOUTH AMERICAN LABIATAE

# EXPLANATION OF PLATE

## PLATE 7

Sphacele intermedia Epling

Ecuador

From the type specimen, *Hitchcock* 20839, in the United States National Herbarium (originally determined as *Sphacele conferta* Bentham).



EPLING SOUTH AMERICAN LABIATAE

# A REVISION OF THE GENUS BOUCHEA (EXCLUSIVE OF CHASCANUM)<sup>1</sup>

#### MYRLE GRENZEBACH<sup>2</sup>

Formerly Teaching Fellow, Missouri Botanical Garden

#### HISTORY

The small genus Bouchea of the Verbenaceae was first described by Chamisso<sup>3</sup> in 1832. The genus as constituted originally embraced two species, namely, B. pseudogervaô, based on specimens collected by Beyrich near Frieburg in Brazil, and B. Ehrenbergii which was described from specimens collected by Ehrenberg near Port au Prince in San Domingo. The former species had been described and illustrated previously by St. Hilaire under the name Verbena pseudogervaô. In 1844 Walpers in his 'Repertorium' recognized the two species of Chamisso and added a third species, B. hyderabadensis, from India.

The next mention of the genus was by Schauer<sup>6</sup> who elaborated the Verbenaceae for De Candolle's 'Prodromus' in 1847. Schauer extended the limitations of the genus Bouchea to include Chascanum Meyer,<sup>7</sup> a small but natural alliance of South African plants. He divided Bouchea into two sections, namely, Rhagocarpium and Chascanum. To the former section he referred six species, four of which—B. pseudogervaô, B. Ehrenbergii, B. laetevirens, and B. agrestis—were attributed to the Western Hemisphere, and two—B. marrubifolia and B. pterygocarpa—to North Africa. To the latter section he referred seven species, six of which—B. cuneifolia, B. cernua, B. garepensis, B. pubescens, B. pinnatifida, and B. adenostachya—are indigenous to South

- <sup>2</sup> Mrs. Lawrence Sherod.
- Cham. in Linnaea 7: 252-254. 1832.
- <sup>4</sup> St. Hil. Pl. Us. des Bres. pp. 1-4, t. 40, 1824-1828.
- <sup>5</sup> Walp. Rep. 4: 11-12. 1844.
- <sup>6</sup> Schauer in DC. Prodr. 11: 557-569. 1847.
- <sup>7</sup> Meyer, Comm. 1: 275–277. 1835.

Issued May 8, 1926.

<sup>&</sup>lt;sup>1</sup> An investigation carried out at the Missouri Botanical Garden in the Graduate Laboratory of the Henry Shaw School of Botany and submitted as a thesis in partial fulfillment of the requirements for the degree of master of science in the Henry Shaw School of Botany of Washington University.

Africa, and one, B. hyderabadensis, to India. Another species, B. pumila of South Africa, was transferred from Chascanum to Bouchea, but being imperfectly known was not given a definite position in either section. Thus fourteen species of Bouchea were recognized by Schauer in De Candolle's 'Prodromus'. Subsequent authors, including Sonders, Gürke, and Pearson, have in general followed Schauer's inclusive generic interpretation of the group.

From 1847 to 1925 additional species of *Bouchea* from America and Africa have from time to time been described, so that the number of species now recorded is more than double the number recognized by Schauer. In the meantime, however, no monographic study or revision of the group has been made.

The present study was undertaken to determine whether Bouchea as amended by Schauer represents a homogeneous and natural genus or whether there might not be at least two distinct elements involved. A careful survey of all species, as far as material could be obtained, has been made and the writer is convinced that Bouchea as circumscribed by Schauer contains two diverse elements which are best regarded as distinct genera. The following revision of the true Bouchea is presented.

## GENERAL MORPHOLOGY

Stems.—The stems in the different species of Bouchea vary from typically herbaceous to distinctly woody and shrubby forms. In some cases the base only is ligneous while others are woody throughout. The stems are sometimes simple, but as a rule they become more or less dichotomously branched. The main axis and the branches may be four-angled or terete. Quadrangular branches are the more common, but the main axis often becomes terete toward the base.

Leaves.—The leaves show considerable diversity in outline, size, texture, and character of margin. In two species the leaves are sessile, while in all others they are petiolate. In the majority

<sup>&</sup>lt;sup>1</sup> Sond. in Linnaea 23: 86. 1850.

<sup>&</sup>lt;sup>2</sup> Gürke in K. Bot. Gart. Berlin Notiz. 3: 74-76. 1900; K. K. Nat. Hofm. Ann. 20: 45. 1905.

<sup>&</sup>lt;sup>3</sup> Pears. in Fl. Cap. 5: 194–207. 1901; S. Afr. Phil. Soc. Trans. 15: 176–180. 1905.

of cases the leaves are more or less ovate, obovate, or subrotund in outline with serrate margins. Incised and entire margins are more infrequent. There are three species in the genus with very distinct foliage. One species has dissected leaves, another has linear leaves with entire margins, while the third species has entire, thick, scabrous, spatulate leaves. These three species can be readily distinguished by their leaf characters. Hence the leaves furnish excellent characters for specific differentiation.

Inflorescence.—The inflorescence is racemose or rarely spicate, commonly terminal or occasionally axillary. The flowers are solitary, mostly short-pedicellate, rarely sessile, subtended by a bract, or by a bract and two bracteoles. The bracts are usually subulate or lanceolate, but in one species, B. spathulata, they are leaf-like. The racemes may be loosely or densely flowered. The character of the inflorescence is comparatively uniform and not of much value in specific determination.

Pubescence.—All the species except one are more or less pubescent, and the pubescence is relatively uniform as to kind. Some species are densely pubescent while others are nearly glabrous. The pubescence in most cases is of short straight hairs. B. agrestis is a notable exception and differs from all other species in having a pubescence of long white, somewhat flaccid hairs. The species can be distinguished by this character.

Calyx.—The calyx, although relatively constant throughout the genus, shows considerable diversity in the different species, and these calyx characters are of use in specific determination in several cases. The calyx is persistent, tubular, five-angled, and five-toothed. There is always one tooth (the posterior lobe) shorter than the other four. Sometimes this difference is very marked, and again it is scarcely noticeable. The calyx varies considerably in texture; some are thin and almost hyaline, while others are of a heavier texture. There is also variation in the length of the teeth.

Corolla.—The corolla is relatively constant throughout the genus, varying chiefly in size and color. The color is usually white, but blue, lilac, and rose-colored flowers are recorded. The corolla is funnel-shaped, somewhat bilabiate, with an elongated tube and a slightly unequal five-lobed, spreading limb.

Stamens.—The stamens are included, didynamous, and inserted on the corolla-tube. The lower pair (antero-lateral) is inserted at about the middle of the tube, opposite the sinuses of the anterior lip. The other pair (postero-lateral) is inserted at a little higher level opposite the sinuses of the posterior lip. The filaments are short. The anthers are ovate to subcordate with two parallel anther-sacs.

Pistil.—The oblong, bilocular, two-ovuled ovary is attached to the receptacle by a short gynophore. The style is long and filiform but included. The stigma is two-lobed. The anterior lobe is the larger and is somewhat subclavate-stigmatose, while the posterior lobe is aborted and tooth-like.

Fruit.—The fruit furnishes the most important characters used in specific determination in the genus. It separates into two distinct cocci at maturity or remains slightly coherent at the base. The cocci are always elongated, more or less beaked, and vary markedly in length. The dorsal surface is convex and usually more or less reticulately ridged. The commissural surface is either plane, ridged, or somewhat furrowed. The beak is very variable in length; it may be conspicuously different in color and texture from the rest of the fruit, and smooth or pubescent, whereas the body of the fruit is striated or reticulately ridged; or it may be quite inconspicuous, noticeable only by a slight contraction of its base. When the beak is greatly differentiated the edges of the fruit are found to have the same texture and surface characteristics as the beak. The fruit may be included in the calyx or exserted.

### GENERIC RELATIONSHIPS

Bouchea belongs to the tribe Verbeneae and is obviously related to Verbena from which it was segregated by Chamisso on account of the separation of the fruit into two, instead of four, nutlets or cocci. It is related also to Stachytarpheta Vahl, but from that genus Bouchea is readily distinguished by the absence of a stout, deeply pitted rachis in which the flowers are more or less immersed. Bouchea is furthermore allied to the genus Priva Adans., particularly through the species P. cuneato-ovatis (Cav.) Rusby, but Priva in nearly all cases has an ampliate-globular, instead of a narrow tubular, fruiting calyx.

The immediate relationship of *Bouchea* is with *Chascanum* Meyer, and the two genera, as stated previously, were united by Schauer. A careful examination of a relatively large series of specimens, however, reveals important morphological differences which may be tabulated as follows:

Bouchea Cham.—Calyx tubular, 5-angled, occasionally slightly cleft at maturity, not inflated; fruit equalling or exceeding the persistent calyx; cocci mostly much longer than broad, distinctly beaked, not usually deeply excavated at the base (pl. 9, figs. 1-12; pl. 10, figs. 13-16; pl. 11, figs. 17-24).

Chascanum Meyer.—Calyx tubular, 5-angled, conspicuously splitting from apex to base at maturity, somewhat inflated; fruit included within the persistent calyx; cocci mostly less than twice as long as broad, not beaked, usually deeply excavated at the base (pl. 11, figs. 25-28).

## GEOGRAPHICAL DISTRIBUTION

The genus *Bouchea* is somewhat limited in its distribution. As here defined, ten species are admitted to the genus and all but one occur in the Western Hemisphere, ranging from New Mexico to Bolivia or between 32° N. and approximately 20° S. The only recognized species of *Bouchea* from the Eastern Hemisphere is *B. pterygocarpa* which is found in Abyssinia.

Three species occur in the United States. B. linifolia is found in southwestern Texas, B. spathulata in western Texas and northern Mexico, while B. prismatica, which is the most widely distributed species, extends southward from New Mexico, through Mexico, Central America, and the West Indies, into Venezuela and Colombia. B. prismatica and B. Nelsonii are the only species known from Central America; the latter species has been collected only in southern Mexico and Guatemala. B. dissecta, the only species limited to Mexico in its distribution, is found in the northwestern part of that country. Three of the four species which are indigenous to South America are, as far as known, rather local in their distribution. B. pseudochascanum occurs in Ecuador, B. agrestis in Brazil, and B. incisa in Bolivia. B. pseudogervaô, however, has a wider distribution. It is recorded from Peru, Bolivia, and Brazil.

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## ABBREVIATIONS

Abbreviations indicating the herbaria where specimens cited herein are deposited are as follows: US = United States National Herbarium; G = Gray Herbarium of Harvard University; M = Missouri Botanical Garden Herbarium; F = Field Museum of Natural History Herbarium; C = University of Chicago Herbarium (at the Field Museum); CC = Columbia College Herbarium (at the New York Botanical Garden).

#### TAXONOMY

Bouchea Cham. in Linnaea 7: 252. 1832; Schauer in DC. Prodr. 11: 557. 1847, excl. *Chascanum*; Mart. Fl. Bras. 9: 197. 1847–1851; Bocq. Rev. Verb. 139. 1861–1863, excl. *Chascanum*; Benth. & Hook. Gen. Pl. 2: 1144. 1873–1874, excl. *Chascanum*; Briq. in Engler & Prantl, Nat. Pflanzenfam. 4<sup>8a</sup>: 153. 1897, excl. *Chascanum*.

Denisaea Neck. Elem. 1: 306. 1790.

Annual or perennial plants, herbaceous to woody, densely pubescent to glabrous. Leaves usually petiolate, sometimes sessile, usually serrate to serrate-crenate, rarely incised, dissected, or entire. Inflorescence racemose, rarely spicate, terminal, seldom axillary, elongate, loosely to densely flowered, bracteate. Flowers solitary, usually pedicellate. Bracts subulate, lanceolate or leaf-like. Calvx persistent, tubular, 5-ribbed, ribs terminating in 5 more or less unequal teeth. Corolla-tube funnelform, cylindrical, erect or curved; limb oblique, spreading, unequally 5-lobed, the two posterior lobes shorter than the anterior lobes. Stamens 4, didynamous, included; filaments short, inserted on the corolla-tube, the posterior pair of stamens inserted at the middle of the tube, the anterior pair inserted at a somewhat higher level: anthers 2-celled, ovate to subcordate. Ovary 2-locular. loculi 1-ovulate, oblong; style filiform; stigma 2-lobed, anterior lobe club-shaped, posterior lobe tooth-like, on a level with the anterior pair of stamens. Fruit dry, linear, beaked, included in the calvx or exserted, separating into two cocci at maturity; cocci totally separate or coherent at the base, dorsal surface more or less reticulately ridged, commissural surface plane, furrowed, or ridged, sometimes a little roughened.

Type species: B. pseudogervaô (St. Hil.) Cham. in Linnaea 7: 253. 1832.

# KEY TO THE SPECIES

- A. Leaves distinctly petiolate; petioles .5-4 mm. in length.
  - - C. Beak oblong, emarginate.
      - D. Leaves ovate to subrotund, dentate to crenate-dentate, or laciniate-dentate.
        - - F. Calyx 5-9 mm. long; beak 1.5 mm. or less in length.
          - - H. Beak 2-3 mm. long, glabrous or slightly pubescent.
    - - I. Plants conspicuously villous-hirsute...... 5. B. agrestis
      - II. Plants not villous-hirsute.
        - J. Fruit exserted.

AA. Leaves sessile.

- 1. B. pterygocarpa Schauer in DC. Prodr. 11: 558. 1847; Engler, Pflanzenw. Ost.-Afr. A. 57 and C. 338. 1895; *ibid.* A. 44. sphalm. *pterygosperma*.

Stem ligneous, 10–15 dm. high, branched; branches somewhat 4-angled, glaucescent; leaves petiolate, ovate to ovate-elliptical, 9–30 mm. long, 7–15 mm. broad, rather thick, somewhat unequally serrate-dentate, obtuse to subacute at the apex, more or less cuneate at the base, scabrous-pubescent on both surfaces; petioles 5–15 mm. long; racemes terminal, subsessile, 8–42 cm. long, pubescent, closely flowered; flowers subsessile; bracts subulate, 2–3 mm. long; calyx 8–9 mm. long, splitting longitudinally from apex to base at maturity, scabrous-pubescent, teeth very short, apex almost truncate; fruit about as long as the calyx, separating into two distinct cocci, dorsal surface slightly ridged, commissural surface with a central longitudinal ridge, roughened, excavated at the base, beak membranous-winged.

Distribution: Abyssinia.

Specimens examined:

Abyssinia: in the mountains near Adeganna, 11 April, 1839, Schimper 1012 (US, M).

- 2. B. prismatica (Jacq.) Kuntze, Rev. Gen. Pl. 2: 502. 1891.
- B. Ehrenbergii Cham. in Linnaea 7: 253. 1832; Walp. Rep. 4: 12. 1844; Torr. in U. S. & Mex. Bound. Surv. 126. 1859; Gray, Syn. Fl. N. Am., ed. 2, 2<sup>1</sup>: 334. 1886; Briq. in Engl. & Prantl, Nat. Pflanzenfam. 4<sup>2a</sup>: 153. 1897.

Verbena prismatica Jacq. Coll. 2: 301. 1788; Icones Pl. Rar. 2: t. 208. 1786-1793.

Zapania prismatica Lam. Encycl. Meth. 1: 59. 1791; Poir. Encycl. Meth. 8: 844. 1808.

Stachytarpheta bifurca Benth. Pl. Hartw. 21. 1839; Walp. Rep. 4:11. 1844.

Stem 1–6 dm. high, 4-angled, more or less pubescent, often furrowed, branched; leaves petiolate, ovate to subrotund, 2–8.5 cm. long, .5–4.5 cm. broad, mucronate-dentate to subcrenate, slightly pubescent on both surfaces, acute to somewhat obtuse at the apex, base entire, cuneate to subtruncate; racemes terminal, 8–25 cm. long, often loosely flowered; flowers small, subsessile; bracts lanceolate, 2–3 mm. long; calyx 7–9 mm. long, teeth nearly 2 mm. long; fruit separating into two distinct cocci, equalling or slightly exceeding the calyx, dorsal surface ridged, commissural surface somewhat furrowed, roughened, beak pronounced, about 1.5 mm. long, straight, emarginate.

Distribution: central and southern Mexico, West Indies to northern South America.

Specimens examined:

Tamaulipas: Tula, 1903, Purpus 485 (US).

Aguascalientes: Aguascalientes, 20 Aug., 1901, Rose & Hay 5949, 6229 (US).

Guanajuato: date lacking, Duges 500 (G).

Vera Cruz: Wartenberg, near Tantoyuca, Prov. of Hausteca, coll. of 1858, *Ervendberg 280* (G).

Puebla: Tehuacan, 1-2 Aug., 1901, Rose & Hay 5949 (US). Oaxaca: Almaloyas, 14 July, 1910, Rusby 49 (US).

Yucatan: Progresso, date lacking, Gaumer 1139, 1160 (F).

Haiti: along roads, Port au Prince, 4 July, 1901, Harshberger 51 (US).

Santo Domingo: Puerto Plata, 26 April, 1906, Raunkiaer 1102 (US); Guayubin, Prov. de Monte Cristi, alt. 100 m. or less, 13-21 Feb., 1921, Abbott 958 (US); roadside, Haina, April, 1921, Faris 189, 199 (US); without locality, Oct., 1909, Türckheim 2532 (F, M, US, G).

Porto Rico: near Coamo siroci los Banos, 11 April, 1885, Sintenis 211 F (F, M, US, G); roadside, Coamo Springs, 22 Nov., 1899, Goll 689 (US); Coamo Springs, 1 July, 1901, Underwood & Griggs 585 (US); Coamo Springs, 24 Nov., 1902, Heller 6109 (F, M, G).

Culebra Island: waste places, Culebra, 3-12 March, 1906, Britton & Wheller 252 (US).

St. Thomas Island: Nov., Eggers 114 (G).

St. Croix Island: east end roadside, 9 June, 1897, Ricksecker 409 (M, F, US).

Curação Island: 15 Nov., 1916, Rose 22012 (US).

Margarita Island: El Valle, 20 July, 1901, Miller & Johnston 205 (M, F, US, G).

Venezuela: between Caracas and La Guayra, alt. 600 m., 16 Sept., 1855, Fendler 853 (G); wet meadows, vicinity of El Valle, near Caracas, 28 Aug., 1921, Pittier 9720 (US, G); on slope near El Zigzag between Caracas and Puerto Cabello, 18 Oct., 1921, Pittier 72 (US); La Trinidad de Maracay, Aragua, alt. 440 m., Jan.—Feb., 1913, Pittier 5830, 5832 (US).

Colombia: open wayside, clay, east of Paso de Caramanta, Cauca Valley, Department of Antioquia, alt. 600-700 m., 20 Sept., 1922, *Pennell 10825* (US).

Colombia: eastern side of Cauca Valley, La Manuelita, near Palmira, Cauca, alt. 1100–1302 m., Dec., 1905–Jan., 1906, *Pittier 833* (US).

# 2a. Var. laciniata Grenzebach, n. var.<sup>1</sup>

Stems like the species, leaves ovate, about 4 cm. long, 1.5-2.5 cm. broad, margins distinctly incised, apex acute to acuminate, base cuneate to subtruncate.

Distribution: east central Mexico.

Specimen examined:

Vera Cruz: near Tantoyuca, Prov. of Huasteca, coll. of 1858, Ervendberg 102 (G, TYPE, photograph in M).

# 2b. Var. brevirostra Grenzebach, n. var.2

Stem, leaf, and raceme characters like the species; calyx 5-7.5 mm. long; fruit about equalling the calyx, or slightly exserted, beak about .5 mm. long, somewhat curved.

- <sup>1</sup> Bouchea prismatica (Jacq.) Kuntze var. laciniata Grenzebach, var. nov., a forma typica recedit foliis ovatis, circiter 4 cm. longis, 1.5–2.5 cm. latis, laciniato-dentatis, acutis vel acuminatis, basi cuneatis vel subtruncatis.—Near Tantoyuca, Province of Huasteca, Vera Cruz, Mexico, coll. of 1858, *Ervendberg 102* (G, TYPE, photograph in M).
- <sup>2</sup> Bouchea prismatica (Jacq.) Kuntze var. brevirostra Grenzebach, var. nov., calyce 5-7.5 mm. longo; fructo calycem subaequanti vel rarius excedenti; rostro circiter 5 mm. longo; aliter formae typicae species simillimum.—Collected at Punguato, vicinity of Morelia, State of Michoacan, Mexico, alt. 2100 m., 9 Aug., 1909, Arsène 2857 (M, TYPE, US).

Distribution: New Mexico, southward to Salvador, also in the Barbados.

Specimens examined:

New Mexico: coll. of 1851-1852, Wright 1508 (US).

Sonora: coll. of 1850-1852, Thurber 1094 (F, G).

Chihuahua: hills and plains near Chihuahua, 2 Sept., 1886, Pringle 994 (M); and Aug.-Sept., 1885, Pringle 325 (G, F); Cerro de Guadeloupe, alt. 2250 m., 3 Sept., 1899, Pringle 7941 (F, G).

Durango: damp, rocky soil, Santiago Papasquiara, Apr. and Aug., 1896, Palmer 416 (US, G, F, M).

San Luis Potosi: region of San Luis Potosi, alt. 1800-2400 m., coll. of 1878, Parry & Palmer 716 (M, G).

Jalisco: Guadalajara, July, 1886, Palmer 261 (G, US).

Colima: Colima, July, 1897, Palmer 104 (US).

Michoacán: Mont. Zacoalco, 10 July, 1865–1866, Bourgeau 545 (US, G); Loma del Zapote, vicinity of Morelia, alt. 1950 m., 25 July, 1912, Arsène 8489 (US); Punguato, vicinity of Morelia, alt. 2000 m., 16 July, 1909, Arsène 3040 (M, G, US); Punguato, vicinity of Morelia, alt. 2100 m., Aug., 1909, Arsène 2857 (M, TYPE, US); Punguato, Morelia, alt. 1950 m., 8 Sept., 1909, Arsène 4 (F).

Guanajuato: coll. of 1909, Furness, without number (F).

Queretaro: near San Juan del Rio, Aug., 1905, Rose, Painter & Rose 9570 (US); locality not indicated, alt. 1850 m., July, 1914, Arsène 9997 (M, US, G).

Mexico: Tlalpam, valley of Mexico, 20 Aug., 1896, Harshberger 152 (G).

Puebla: vicinity of San Luis Tultitlanapa, near Oaxaca, June, 1908, Purpus 3405 (F, M, US, G).

Oaxaca: valley of Etta, Sept., 1895, Alvarez 747 (G).

Guatemala: Santa Rosa, Department of Santa Rosa, alt. 900 m., June, 1892, Smith 2965 (US, G).

# 2c. Var. longirostra Grenzebach, n. var. 1

<sup>&</sup>lt;sup>1</sup> Bouchea prismatica (Jacq.) Kuntze var. longirostra Grenzebach, var. nov., calyce 7.5–10 mm. longo; fructo 9–11 mm. longo, rostro erecto, 2–3 mm. longo, exserto.—Collected along Hope Road, Jamaica, alt. 120 m., 14 Nov., 1914, Harris 11792 (M, TYPE, F, G).

Salvador: dry slope, vicinity of San Vincente, Department of San Vincente, alt. 350-500 m., 2-11 March, 1922, Standley 21620 (US).

Stem and leaf characters like the species; calyx 7.5-10 mm. long; fruit 9-11 mm. long, beak 2-3 mm. long, straight, exserted.

Distribution: southern Mexico, Bahamas and West Indies to northern South America.

Specimens examined:

Oaxaca: Cuicatlan, 15 July, 1895, Smith 411 (G); vicinity of Cuicatlan, alt. 540-750 m., 8-24 Oct., 1894, Nelson 1597 (US). Yucatan: 17 March, 1903, Seler 3957 (F, G).

New Providence: waste ground, Fort Charlotte, 14 Sept., 1904, Britton & Brace 782 (F).

Cat Island: waste lands, the Bight and vicinity, 1-6 March, 1907, Britton & Millspaugh 5796 (F).

Cuba: damp ground, Havana, 11 May, 190-, Curtiss & West, without number (F); Cienegnith, district of Cienfuegos, Prov. of Santa Clara, 17 June, 1895, Combs 154 (F, G, M, C); waste grounds, vicinity of Tiffin, Camaguey, 14-17 Oct., 1909, Shafer 2861 (US); in orange grove, valley of Rio Matamoras, south of Halguin, Oriente, 14 April, 1909, Shafer 1364 (F); Santiago de las Vegas, 15-20 March, 1905, Hitchcock, without number (F); Santiago de las Vegas, 30 June, 1904, Baker & Wilson 524 (F, US); low ground, Tueabanda, 21 May, Wright 3660 (US).

Jamaica: Hope Road, alt. 120 m., 14 Nov., 1914, Harris 11792 (M, TYPE, F, G); Port Royal, 18 Dec., 1890, Hitchcock, without number (M); streets of Kingston, 9 Dec., 1890, Hitchcock, without number (M); along the railroad between Kingston and Gregory Park, sea level, 22 Feb., 1920, Maxon & Killip 314 (US); exact locality not indicated, coll. of 1850, Alexander, without number (US).

Haiti: open waste places, vicinity of Pikmi, Gonave Island, 5-9 July, 1920, Leonard 5219 (US); in cultivated fields, vicinity of St. Marc, near sea level, 25-28 Feb., 1920, Leonard 2981 (G, US); vicinity of Port au Prince, 21-23 Feb., 1920, Leonard 2852 (US).

Porto Rico: limestone, La Vigia Ponce, 14 March, 1915, Britton, Cowell & Brown 5378 (F, M).

Venezuela: in savannas or in wooded gorges, lower Cotiza, near Caracas, alt. 800-1200 m., June, 1918, Pittier 7887 (US).

# 3. B. Nelsonii Grenzebach, n. sp.<sup>1</sup>

Herbaceous, more or less pubescent throughout, especially above; stems 2.5–6 dm. high, terete below, 4-angled and furrowed above, sparingly branched; leaves petiolate, ovate to subrotund, 2–6 cm. long, 1–4.5 cm. broad, mucronate-dentate, acute to obtuse at the apex, narrowed slightly into the petiole or almost truncate at the base, pubescent on both surfaces, especially along the nerves; inflorescence racemose, terminal or axillary, usually densely flowered, 10–15 cm. long, .8–1 cm. broad; flowers short-pedicellate; bracts linear-lanceolate, about 5 mm. long, pubescent; calyx erect and narrow, 13–15 mm. long, pubescent; fruit separating into two distinct cocci at maturity, 11–16 mm. long, dorsal surface somewhat ridged, commissural surface plane, a little rough, beak about one-third the length of the entire fruit, 4–6 mm. long, slightly pubescent at the tip.

Distribution: southern Mexico and Guatemala.

Specimens examined:

Oaxaca and Chiapas: between Topana, Oaxaca, and Tonala, Chiapas, alt. 60–150 m., 1–3 Aug., 1895, Nelson 2867 (US, TYPE, G, photograph and fragments in M).

Guatemala: Zacapa, alt. 180 m., 24 Jan., 1905, Deam 173 (G), slender form.

This species resembles B. prismatica (Jacq.) Kuntze to which the specimens cited have been referred hitherto, but it differs in having longer fruit, with a distinctly longer and pubescent beak,

<sup>1</sup> Bouchea Nelsonii Grenzebach, sp. nov., herbacea plus minusve pubescens; caulibus 2.5-6 dm. altis inferne teretibus superne quadrangularibus sulcatisque, parce ramosis; foliis petiolatis, ovatis vel subrotundatis, 2-6 cm. longis, 1-4.5 cm, latis, mucronato-dentatis, acutis vel obtusis, basi cuneatis vel subtruncatis, utrinque pubescentibus; inflorescentiis racemosis, terminalibus vel axillaribus racemis 10-15 cm. longis, .8-1 cm. latis; floribus crebre brevi-pedicellatis; bracteis lineari-lanceolatis, circiter 5 mm. longis, hirtellis; calyce erecto, plicato-angulato, 13-15 mm. longo, hirtello, dentibus 5 subulatis, inacqualibus; fructo exserto maturitate in 2 distincte cocci sponte secedens, coccis linearibus, 11-16 mm. longis, dorso striatis vel parce reticulato-jugis, commissura plana verruculosa, rostro 4-6 mm. longo ad apicem parce pubescens.—Between Topano, Oaxaca and Tonala, Chiapis. Mexico, alt. 60-150 m., Aug. 1-3, 1895, Nelson 2367 (US, TYPE, G, photograph and fragments in M).

longer and pubescent calyx, and usually a stouter, denser, and broader inflorescence. The entire plant, furthermore, is more pubescent than *B. prismatica*.

## 4. B. dissecta Wats. in Proc. Am. Acad. 24: 68. 1889.

An annual, distinctly herbaceous, slender, very finely puberulent to glabrous; stems 4–6.5 dm. high, 4-angled, sulcate; leaves ovate, 2–7 cm. long, 1–4 cm. broad, thin, pinnately cleft nearly to the midrib, the narrow segments entire, or 1–3-toothed, minutely pubescent; racemes terminal, 10–30 cm. long, slender, loosely flowered; flowers short-pedicellate; bracts subulate, only a little longer than the pedicels; calyx 7–8 mm. long, shortly toothed, slightly pubescent, thin; corolla white; fruit 10–12 mm. long, about one-third longer than the calyx, conspicuously long-beaked, beak 3.5–4 mm. long, dorsal surface longitudinally ridged, commissural surface somewhat furrowed, smooth.

Distribution: northwestern Mexico.

Specimens examined:

Sonora: rocky ridges, Guaymas, Oct., 1887, Palmer 259 (G, TYPE); Agiabampo, 3-5 Oct., 1890, Palmer B (G).

Sinaloa: Culiacan, 27 Aug.-15 Sept., 1891, Palmer 1485 (G, US); San Augustin, San Ignacio, coll. of 1921, Ortega 621 (US).

# 5. B. agrestis Schauer in DC. Prodr. 11: 558. 1847, and in Mart. Fl. Bras. 9: 197. 1847-1851.

An annual, villous-hirsute in the younger stages, more or less glabrate; branches somewhat 4-angled; leaves short-petiolate, obovate, elliptical-oblong, 1.5-4 cm. long, 1-1.7 cm. broad, acutely serrate from the middle of the leaf to the apex, entire towards the base, attenuate on the petiole, villous-hirsute; racemes terminal, slender, loosely flowered; flowers pedicellate; bracts linear, 5-6 mm. long; calyx 7-9 mm. long, hirsute, teeth long; corolla lilac to rose; fruit separating into two distinct cocci, 6.5-8 mm. long, included within the calyx, beak long, attenuate, slightly pubescent, dorsal surface distinctly ridged, commissural surface plane, smooth.

Distribution: Brazil. Specimen examined:

Brazil: vicinity of Bahia, date lacking, Blanchet 3731 (M).

6. B. pseudogervaô (St. Hil.) Cham. in Linnaea 7: 253. 1832; Walp. Rep. 4: 11. 1844; Schauer in DC. Prodr. 11: 557. 1847; and in Mart. Fl. Bras. 9: 195. 1847-1851.

Verbena pseudogervaô St. Hil. Pl. Us. des Bres. pp. 1–4. t. 40. 1824–1828.

(?) Verbena fluminensis Vellozo, Fl. Flum., t. 38. 1827.

Stem 6–9 dm. high, somewhat ligneous, stout, almost glabrous, below terete; branches usually 4-angled, glabrous to slightly pubescent; leaves petiolate, ovate to elliptical-oblong, 6–10 cm. long, 2.5–5 cm. broad, membranous, coarsely mucronate-dentate, acuminate, entire and cuneate at the base, essentially glabrous on both surfaces, dark green above, pale beneath; inflorescence racemose, terminal, 10–30 cm. long, glabrous or slightly pubescent; flowers short-pedicellate, almost sessile; bracts linear-lanceolate, about 5 mm. long; bracteoles about one-third as long as the bracts; calyx 10–13 mm. long, finely pubescent; fruit of two cocci coherent at the base, cocci almost cylindrical, slightly exserted beyond the calyx, beak short, obscure, only slightly contracted at the base, dorsal surface ridged from base to apex, commissural surface plane or slightly convex, smooth.

Distribution: Brazil. Specimens examined:

Peru: in hedge-rows, La Merced, 19-24 Aug., 1923, Macbride 5304 (F).

Bolivia: Junction of Rivers Beni and Madre de Dias, Aug., 1886, Rusby 915 (F, M, US, G); near Cochabamba, 1891, Bang 2001 (CC).

Brazil: Minas Geraes, 31 Oct., 1856. Regnell 340 (US).

- 7. B. pseudochascanum (Walp.) Grenzebach, n. comb.
- B. laetevirens Schauer<sup>3</sup> in DC. Prodr. 11: 557. 1847, and in Mart. Fl. Bras. 9: 196. 1847-1851.
- <sup>1</sup> Examination of specimens of *B. pseudogervaô* from Bolivia and Peru show them to have a slightly longer and more attenuate beak than the specimens studied from Brazil, but this difference is not great enough in the material at hand to warrant even varietal differentiation.
- <sup>3</sup> Although Vellozo used the specific name *fluminensis* in referring to this species in his 'Flora Fluminensis' in 1827, yet the illustration is unaccompanied by a description, and it seems advisable, therefore, to retain the name *pseudogervað*.
  - It is impossible to separate B. laetevirens and B incrassata specifically. The

(?) B. incrassata Lange, Ind. Sem. Hort. Haun. 31. 1870; Bot. Tidssk. 8: 3. 1874-1876.

Stachytarpheta pseudochascanum Walp. Rep. 4:11. 1844.

Stems somewhat ligneous, terete, glabrous at the base; branches obtusely 4-angled, erect-spreading, pubescent; leaves short-petiolate, ovate to subrotund or elliptical-ovate, 2–7 cm. long, 1.5–3 cm. broad, serrate, acute to subobtuse, entire, cuneate at the base, young leaves pubescent on both surfaces, glabrate except along the nerves beneath; petioles 6–12 mm. long; inflorescence racemose, terminal or axillary, 14–30 cm. long, pubescent; flowers short-pedicellate; bracts subulate, short, a little longer than the pedicels; bracteoles minute; calyx about 8–9 mm. long, almost truncate at the apex, teeth very short, triangular, slightly pubescent, ciliate, occasionally splitting along one side; fruit separating into two cocci at maturity except at the slightly coherent base, about one-third longer than the calyx, beak short, attenuate, dorsal surface ridged, commissural surface plane, almost smooth.

Distribution: Ecuador. Specimens examined:

Ecuador: Caraques, 23 June, 1923, Anthony & Tate 87 (US).

8. B. incisa Rusby in Bull. N. Y. Bot. Gard. 4: 432. 1907. Stem somewhat ligneous, glabrous to slightly pubescent, terete below, purplish, finely striate, branched; branches somewhat 4-angled; leaves short-petiolate, ovate, 5-12 cm. long, 2-4 cm. broad, upper half somewhat incisely serrate toward the apex or rarely entire, acuminate, entire at the apex and base, glabrous or slightly pubescent on both surfaces, especially along the nerves on the under side, green above, pale beneath; racemes terminal, 1-3 dm. long; flowers shortly and stoutly pedicellate; bracts about 3 mm. long, subulate, pubescent; bracteoles about one-third as long as the bracts; calyx about 1.5 cm. long, pubescent, cylindrical, recurved in anthesis, erect in fruit; corolla-tube nearly 2 cm. long, strongly recurved, limb broad; fruit about 1.5 cm. long, the two cocci slightly coherent at the base, beak short,

descriptions of the two are practically the same, and the excellent illustrations in Bot. Tidssk. 8: t. 2. 1874–1876, and in Mart. Fl. Bras. 9: t. 33. 1847–1851, show them to be the same in all essential details.

rather inconspicuous, dorsal surface slightly ridged, commissural surface plane, smooth.

Distribution: Bolivia.

Specimens examined:

Bolivia: without exact locality and date of collection, Bang 2226 (CC, TYPE M, G, F).

B. linifolia Gray in Am. Jour. Sci. II. 16:98. 1853; Torr. in U. S. & Mex. Bound. Surv. 2:126. 1859; Gray, Syn. Fl., ed. 2, 2:335. 1886; Coult. Bot. Western Texas, 326. 1891–1894.

Stem simple or fastigiately branched from a somewhat woody base, 3–6 dm. high, glabrous; branches rigid, striate, sulcate, very leafy; leaves sessile or nearly so, linear to linear-lanceolate, 2–4.5 cm. long, 2–5 cm. broad, acute at both ends; racemes terminal or axillary, 4–15 cm. long, loosely flowered; pedicels about 2 mm. long; bracts linear to linear-lanceolate, 2–3 mm. long, somewhat longer than the pedicels; calyx 10–13 mm. long, slender, glabrous; corolla large, limb wide-spreading; fruit separating into two distinct cocci, barely included in the calyx, pubescent along the margin, dorsal surface ridged, commissural surface smooth or nearly so, beak pointed, villous.

Distribution: western and southern Texas.

Specimens examined:

Texas: west Texas to El Paso, New Mexico, May-Oct., 1849. Wright 449 (US); valley of the Rio Grande below Donana, date lacking, Emory 814 (US); dry calcareous hillsides, Montell, Uvalde County, 15 Oct., 1917, Palmer 13007 (M); Neuces River, date lacking, Havard 1383 (M); San Pedro, coll. of 1851-1852, Wright 1509 (M, US).

10. B. spathulata Torr. in U. S. & Mex. Bound. Surv. 2: 126. 1859; Gray, Syn. Fl., ed. 2, 2<sup>1</sup>: 335. 1886; Coult. Bot. Western Texas, 326. 1891–1894.

Distinctly ligneous, 3-6 dm. high, usually branched; branches terete, softly pubescent, very leafy; leaves sessile, obovate, spatulate, 5-18 mm. long, 3-7 mm. broad, entire, obtuse, acute at the base, coriaceous, scabrous; spikes terminal, short, loosely flowered; flowers divergent from the rachis; bracts leaf-like, oblanceolate, about three-fourths the length of the calyx; calyx

8-11 mm. long, scabrous-pubescent; corolla much exceeding the calyx; fruit separating into two distinct cocci at maturity, not exserted above the calyx, dorsal and commissural surfaces smooth, beak pointed, pubescent, margins of the fruit also pubescent.

Distribution: western Texas and northern Mexico.

Specimens examined:

Texas: mountains east of Tornillo Creek, Aug., 1883, *Havard 96* (US); Canyon Boquillas, 3 Aug., 1919, *Hanson 718* (US).

Coahuila: Sierra de la Poila, Oct., 1910, Purpus 4750 (F, M, G).

## LIST OF EXCLUDED SPECIES

Bouchea adenostachya Schauer in DC. Prodr. 11: 560. 1847 = Chascanum.

- B. caespitosa Pearson in Trans. S. Afr. Phil. Soc. 15: 178. 1904 = Chascanum.
- B. cernua Schauer in DC. Prodr. 11:559. 1847 = Chascanum cernuum Meyer, Comm. 1:276. 1897.
- B. copiapensis Gay, Hist. Chile 5:26. 1849 = Priva cuneato-ovata (Cav.) Rusby.
- B. cuneifolia Schauer in DC. Prodr. 11: 559. 1847 = Chascanum cuneifolium Meyer, Comm. 1: 276. 1897.
- B. garepensis Schauer in DC. Prodr. 11: 560. 1847 = Chascanum garepense Meyer, Comm. 1: 277. 1897.
- B. glandulifera Pearson in Fl. Cap. 5: 204. 1901. = Chascanum.
  - B. Hanningtonii Oliver in Hook. Ic. Pl. t. 1446 = Chascanum.
  - B. hederacea Sond. in Linnaea 23:86. 1850 = Chascanum.
- B. incisa Pearson in Trans. S. Afr. Phil. Soc. 15: 180. 1904 = Chascanum.
- B. integrifolia Pearson in Trans. S. Afr. Phil. Soc. 15: 179. 1904 = Chascanum.
- B. Krookii Guerke in Ann. Nat. Hofmus. 20: 45. 1905 = Chascanum.
  - B. latifolia Harv. Thes. Cap. 2: 57. = Chascanum.
- B. longipetala Pearson in Fl. Cap. 5: 199. 1901 = Chascanum.
- B. marrubiifolia Schauer in DC. Prodr. 11: 558. 1847 = Chascanum.

- B. namaquana Bolus, ex Pearson in Fl. Cap. 5: 204. 1901 = Chascanum.
- B. pinnatifida Schauer in DC. Prodr. 11: 560. 1847 = Chascanum pinnatifidum Meyer, Comm. 1: 277. 1897.
- B. pumila Schauer in DC. Prodr. 11:500. 1847 = Chascanum pumilum Meyer, Comm. 1:277. 1897.
- B. rariflora Chiov. in Ann. Bot. Roma 9: 127. 1911 = Chascanum.
- B. Schlechteri Guerke in Notiz. K. Bot. Gart. Berlin 3: 75. 1903 = Chascanum.
- B. sessilifolia Vatke in Linnaea 43: 529. 1880-1882 = Chascanum.
- B. Wilmsii Guerke in Notiz. K. Bot. Gart. Berlin 3: 74. 1903 = Chascanum.

## DOUBTFUL SPECIES

B. hyderabadensis Walp. Rep. 4: 12. 1844, is a species not sufficiently known for definite specific determination.

## LIST OF EXSICCATAE CITED

Distribution numbers are in *italics*. The numbers in parentheses are those of the species in the present revision. Collections distributed without numbers are indicated by a dash.

Abbott, W. L. 958 (2).
Alexander, R. C.—(2c).
Alvarez, C. 747 (2b).
Anthony, H. E. & Tate, G. H. H. 87 (7).
Arsène, Bro. G. 4, 2857, 3040, 8489, 9997 (2b).
Baker, C. E. & Wilson, 524 (2c).
Bang, A. M. 2001 (6a); 2226 (8).
Blanchet, J. S. 3731 (5).
Bourgeau, E. 545 (2b).
Britton, N. L. & Brace, L. J. K. 782 (2c).
Britton, N. L., Cowell, J. F. & Brown, S. 5378 (2c).

Britton, N. L. & Millspaugh, C. F. 5796 (2c).
Britton, N. L. & Wheller, W. M. 252 (2).
Combs, R. 154 (2c).
Curtiss & West,—(2c).
Deam, C. C. 173 (3).

Dugès, A. διν (၁) Eggers, Baron H. F. A. 111 (2). Emory, W. H. 814 (9). Ervendberg, L. C. 102 (2a); 280 (2). Faris, J. A. 189, 199 (2). Fendler, A. 853 (2). Furness, D. R.—(2b). Gaumer, G. F. 1139, 1160 (2). Goll, G. P. 689 (2). Hanson, H. C. 718 (10). Harris, Wm. 11792 (2c). Harshberger, J. W. 51 (2); 152 (2b). Havard, V. 96 (10); 1383 (9). Heller, A. A. 6109 (2). Hitchcock, A. S.—(2c). Leonard, E. C. 2852, 2981, 5219 (2c). Macbride, J. F. 5304 (6).

Maxon, W. P. & Killip, E. P. 314 (2c).

Miller, O. O. & Johnston, J. R. 205 (2). Nelson, E. W. 1597 (2c); 2807 (3).

Ortega, J. G. 621 (4).	Rose, J. N., Painter, J. H. & Rose, J. S.
Palmer, E. 104, 261, 416, (2b); B, 259,	9570 (2b).
1485 (4).	Rusby, H. H. 49 (2); 915 (6a).
Palmer, E. J. 13007 (9).	Schimper, W. 1012 (1).
Parry, C. C. & Palmer, Ed. 716 (2b).	Schumann, W. 232 (2).
Pennell, F. W. 10825 (2).	Seler, Caec. and E. E. 3957 (2c).
Pittier, E. 72 (2).	Shafer, J. A. 1364, 2861 (2c).
Pittier, H. 833, 5830, 5832, 9720 (2);	Sintenis, P. 211F (2).
7887 (2c).	Smith, J. D. 2965 (2b).
Pringle, C. G. 325, 994, 7941 (2b).	Smith. L. C. 411 (2c).
Purpus, C. A. 485 (2); 3405 (2b); 4750	Standley, P. C. 21620 (2b).
(10).	Thurber, G. 1094 (2b).
Raunkiaer, C. 1102 (2).	Underwood, L. M. & Griggs, R. F. 585
Regnell, A. A. 340 (6).	(2).
Ricksecker, Mrs. Rev. J. J. 409 (2).	von Türckheim, H. 2532 (2).
Rose, Mr. & Mrs. J. N. 22012 (2).	Wright, C. 449, 1509 (9); 1508, (2b);
Rose, J. N. & Hay, R. 5949, 6229 (2).	3660 (2c).

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New species, varieties, and combinations are printed in **bold face** type; synonyms in *italics*; and previously published names in ordinary type.

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incisa		Denisaea
incrassata · · · · · · · · · · · · · · · · · ·		Stachytarpheta
alelevirens		bifurca
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## EXPLANATION OF PLATE

#### PLATE 8

Geographical distribution of the genus Bouchea.

The generic distribution of the genus *Bouchea* is shown by the outlined areas. The specific distribution is indicated by numerals which correspond to the numbers of the various species as treated in this revision.

1. B. pterygocarpa

2. B. prismatica

3. B. Nelsonii

4. B. dissecta

5. B. agrestis

6. B. pseudogervaô

7. B. pseudochascanum

8. B. incisa

9. B. linifolia

10. B. spathulata

CHANZER VIEW FLATER OF ROTHER

#### EXPLANATION OF PLATE

#### PLATE 9

#### Bouchea prismatica (Jacq.) Kuntze

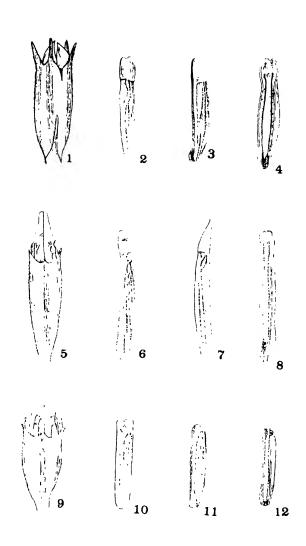
- Fig. 1. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 2. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 3. Mature coccus, side view,  $\times$  5.
- Fig. 4. Mature coccus, commissural surface,  $\times$  5.

#### Bouchea prismatica var. longirostra

- Fig. 5. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 6. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 7. Mature coccus, side view,  $\times$  5.
- Fig. 8. Mature coccus, commissural surface,  $\times$  5.

#### Bouchea prismatica var. brevirostra

- Fig. 9. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 10. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 11. Mature coccus, side view,  $\times$  5.
- Fig. 12. Mature coccus, commissural surface, × 5.



GRENZEBACH REVISION OF BOUCHEA

#### EXPLANATION OF PLATE

#### PLATE 10

#### A

# Bouchea Nelsonii Grenzebach

#### Southern Mexico and Guatemala

From the type specimen, Nelson 2867, in the United States National Herbarium.

В

- Fig. 13. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 14. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 15. Mature coccus, side view,  $\times$  5.
- Fig. 16. Mature coccus, commissural surface,  $\times$  5.











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#### EXPLANATION OF PLATE

#### PLATE 11

#### Bouchea pseudogervað (St. Hilaire) Cham.

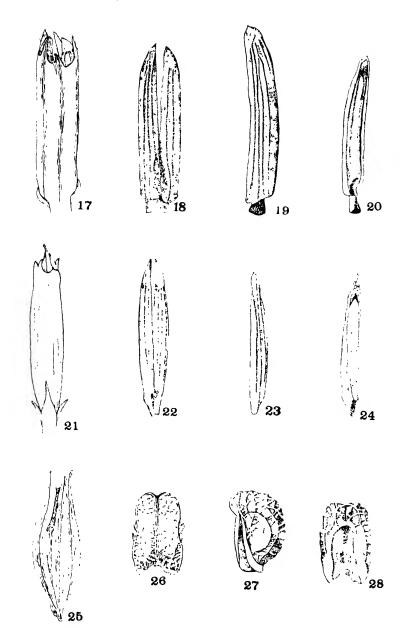
- Fig. 17. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 18. Mature cocci, side view,  $\times$  5.
- Fig. 19. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 20. Mature coccus, commissural surface,  $\times$  5.

# Bouchea pseudogerva® (St. Hilaire) Cham. (showing the more attenuate character of the beak of the fruit)

- Fig. 21. Mature cocci within persistent calyx,  $\times$  5.
- Fig. 22. Mature cocci, side view,  $\times$  5.
- Fig. 23. Mature coccus, dorsal surface,  $\times$  5.
- Fig. 24. Mature coccus, commissural surface, × 5.

#### Chascanum cernuum Meyer

- Fig. 25. Mature fruit within persistent calyx,  $\times$  5.
- Fig. 26. Mature fruit, dorsal surface,  $\times$  5.
- Fig. 27. Mature fruit, side view,  $\times$  5.
- Fig. 28. Mature fruit, commissural surface,  $\times$  5.



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#### EXPLANATION OF PLATE

#### PLATE 12

#### Open calyces.

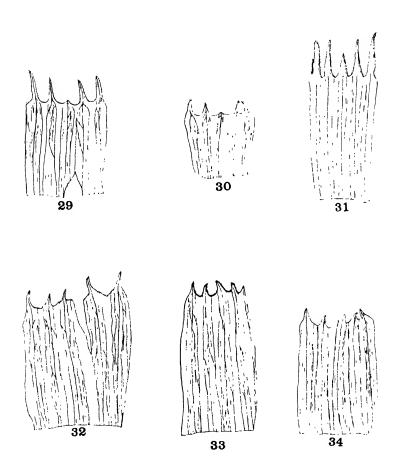
Fig. 29. B. prismatica (Jacq.) Kuntze,  $\times$  5.

Fig. 30. B. prismatica var. brevirostra,  $\times$  5.

Fig. 31. B. Nelsonii Grenzebach, × 5.

Figs. 32, 33. B. pseudogerval (St. Hil.) Cham., × 5.

Fig. 34. Chascanum cernuum Meyer, × 5.



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# FURTHER STUDIES ON THE SUBTERRANEAN ALGAL FLORA OF THE MISSOURI BOTANICAL GARDEN

#### GEORGE T. MOORE

Director of the Missouri Botanical Garden Engelmann Professor in the Henry Shaw School of Botany of Washington University

#### AND

#### NELLIE CARTER

Formerly Research Assistant, Missouri Botanical Garden

## HISTORICAL INTRODUCTION

Although it has long been recognized that the surface of soil forms a very suitable habitat for many algae, especially the Cyanophyceae, it is only in comparatively recent times that investigators have realized that these small plants may possibly play an important part in the biology of the soil. The history of the investigation of soil algae is therefore of comparatively recent date, and, as contrasted with that of the other soil organisms, such as bacteria or protozoa, our knowledge of the algal flora of soil is very imperfect. It seems possible that the presence of these autotrophic plants in the soil may be of great importance, and that their physiological processes may greatly influence soil conditions and also have important effects on the soil as a medium of growth for the other soil organisms. However, any such influence exerted by soil algae is yet to be proved, for we are still in complete ignorance on this matter.

The literature bearing on the problem of soil algae, either directly or indirectly, is nevertheless very extensive although confined to two or three main lines of investigation, namely, the

systematic study of soil algae, their relation to nitrogen, whether or not they are capable of fixing atmospheric nitrogen, and lastly, an immense amount of work has been done on the physiology of algae, their relation to light and organic food substances, which is very important in the soil algae question. Since no very complete review of this literature exists, it is desirable that a summary of the more important papers be made here. The three phases of the literature mentioned above will be taken up separately.

#### THE SYSTEMATIC STUDY OF SOIL ALGAE

Although the systematic examination of the algae occurring in or on the surface of the soil was not undertaken for many years after the development of the soil algae problem, it will possibly be best to begin with this phase in order that some idea may be obtained of the nature of the organisms concerned.

The first extensive work relating to the soil algal flora was published by Esmarch ('10) and dealt with samples taken chiefly from soil in German African colonies. Esmarch's method of culturing his soil samples was such as to favor the growth of Cyanophyceae, and he confined himself almost exclusively to the consideration of these forms. The work first published by him was not undertaken primarily as an investigation of soil algae, but rather to increase the knowledge of the distribution of the Cyanophyceae. The soil samples fell into two main groups as regards the depth from which they were taken, namely, 1-25 cm. and 25-50 cm. Altogether, between 30 and 40 species were identified, chiefly species of the Oscillatoriaceae and Nostocaceae. In considering his data Esmarch came to the conclusion that the majority of the species occurred in the samples from the upper 25 cm., and that a greater proportion of the samples from the lower depths produced no growth. He was stimulated by these results to take up in greater detail the study of soil algae, with the result that in 1914 he published a second paper dealing with extensive investigations of the soil algae in Germany. Esmarch's second paper deals chiefly with the relation of the soil algal flora to depth and to cultivation. With regard to cultivation, surface samples taken from cultivated soils almost always produced a good growth of algae, whereas very few samples from uncultivated soils produced algae except damp sandy soil from the shores of the river or lakes. At 10-50 cm. below the surface no algae could be obtained from soil samples from uncultivated soils, while samples from a similar depth in cultivated soils frequently produced a growth, though fewer species were represented than at the surface. Esmarch felt that since in cultivated soils the lower strata contained approximately the same species as the surface, with the exception that fewer species were present, the relation between the two levels might be such that the lower layers derive their flora directly from the surface in the operations of cultivation, such as ploughing or by the action of earthworms or of seepage water. Esmarch experimented also on the effect of prolonged darkness on some of the algae isolated by him from the soil, but was unable to produce any conclusive proof that they are able to persist for any great length of time in total darkness.

Robbins ('12), investigating the algal flora of the surface and first few inches of soils in Colorado with special reference to their extraordinary nitrogen-fixing capacity, in spite of their low content of organic matter, recognized about 21 species of algae, chiefly Cyanophyceae, but including two Chlorophyceae and a diatom. He believed that the abundant blue-green algae formed a source of carbohydrate food for the nitrogen-fixing organism, Azotobacter, and that for this reason the bacteria were able to flourish in quantity in spite of the low organic content of the soil.

Petersen's work in 1915 deals chiefly with diatoms growing on the surface of the soil, but some attention is also given to *Chloro*phyceae, although the *Cyanophyceae* are entirely ignored.

Bristol ('19a) gives an interesting account of algae obtained from soil samples preserved in an air-dry condition for many years and extended our knowledge not only of the degree of resistance to desiccation of these forms, but also the range of species of the soil algal flora. Two years later the same author (Bristol, '21) published a more extensive soil flora. From an investigation of 50 soil samples taken from the surface 6 inches of soil she identified 20 species of *Chlorophyceae*, 24 *Cyanophyceae*, and 20 diatoms. This must be regarded as the most complete algal flora of the soil published so far. The *Chlorophyceae* included chiefly unicellular

forms, although Bumilleria exilis and Ulothrix subtilis were almost constantly present; the Cyanophyceae were largely species of Nostoc, Anabaena, and Phormidium, and the diatoms, Navicula and Nitzschia spp.

Moore and Karrer ('19), taking up again the investigation of the lower strata of the soil as initiated by Esmarch, established the existence of diatoms and other algae at depths up to 100 cm. below the surface of the ground, the greatest depth at which such organisms had been recorded. The interesting fact was discovered that a unicellular green alga which they identified as *Protoderma viride*<sup>1</sup> is constantly present as far down as 1 m. below the surface. Repeating Esmarch's subterranean culture method, in a slightly varied form, they found that this algae could live and remain green when sunk into the ground for a period of 4–5 months.

#### THE RELATION TO ATMOSPHERIC NITROGEN

By far the greatest amount of work done so far has been concentrated on the relation of the algae to nitrogen and especially to atmospheric nitrogen.

Even in the early fifties following the work of Boussingault and his contemporaries on the nitrogen relations of the higher plants, Laurent ('54) and Morren ('54) came independently to the conclusion that lower organisms, including protozoa and algae, are unable to make use of atmospheric nitrogen, but pass into a resting condition when the supply of combined nitrogen becomes depleted in the medium in which they are living.

For more than 30 years no further work seems to have been done until the opposite opinion was put forward by Frank ('88), who discovered that if soil is kept moist with distilled water there is an increase in the nitrogen content after standing for several months. Frank's attention was drawn to the fact that the increase in nitrogen was not present in the form of nitrates, but in an organic form. Furthermore, on the surface of the soil he noticed that a mat of algae had developed, including Oscillatoria spp., Chlorococcum humicola, and others. Frank thus came to the conclusion that atmospheric nitrogen is transformed into nitrates

<sup>&</sup>lt;sup>1</sup> This now appears to be identical with *Chlorococcum humicola* (Näg.) Rab. as recorded by Bristol ('19b, '21) and others.

by the algae in the soil, and that these are later elaborated into a complex organic form.

In the same year Gautier and Drouin ('88), working with soils containing nitrogen only in an ammoniacal form, found that such soils, if poor in organic matter, show a lower nitrogen content after standing, but that a higher nitrogen content will result if much organic matter is present. In all cases, however, irrespective of the organic content of the soil, the ammoniacal nitrogen decreases and the organic nitrogen increases, the latter in direct proportion to the strength of the algal mat which develops on standing. These workers therefore drew the somewhat original conclusion that the algae are important as conservers of ammoniacal nitrogen, whose escape from the soil they prevent, rather than as fixers of atmospheric nitrogen. They thought that the escaping ammoniacal nitrogen is absorbed by the algae and transformed into a more stable organic form.

In the following year Frank ('89) put forward what he considered to be direct proof of the nitrogen-fixing power of the lower algae by demonstrating that whereas a substantial increase in nitrogen content takes place if soil is kept moist and exposed to daylight, no increase takes place in the same soil if it is kept in the dark, or sterilized. Either of these last two treatments prevents the growth of algae, while the darkness, although excluding the algae, permits growth of bacteria. Frank believed, therefore, that the increase of nitrogen taking place in the light was due entirely to the growth of algae.

The work of Schloesing and Laurent ('92) lent further support to Frank's idea. These workers used poor subsoil and sand for their substratum, and inoculated with impure suspensions of algae, adding also a small sample of ordinary soil extract. They kept their soil in a confined atmosphere and had an arrangement for determining the actual change in the gaseous nitrogen contained in that atmosphere. In this way they were able to check up the loss of gaseous nitrogen with the amount of nitrogen increase in the soil. With Nostoc punctiforme and Cylindrospermum majus they found a substantial increase in the nitrogen content of the soil, and they also established the fact that there is a greater increase of nitrogen in the upper layer of the soil con-

taining the algae than lower down. While realizing that bacteria possibly play a part in the increase of nitrogen, they believed that the algae, being present in larger numbers, are responsible in a greater measure for the fixation of atmospheric nitrogen which occurs.

In 1893, Koch and Kossowitsch kept sand cultures inoculated with a suspension of algae in daylight, and similar cultures as controls in darkness. Their experiments served to confirm the conclusions of Frank that in the presence of light active fixation of atmospheric nitrogen takes place, but that if the growth of algae is prevented by absence of light no fixation occurs.

Since this time there have been repeated investigations of the activities of algae in association with soil bacteria with special reference to nitrogen relations. One of the most energetic workers in this line was Bouilhac. In 1896, claiming to have isolated in pure culture certain algae, this worker found that Nostoc punctiforme, Schizothrix lardacea, and Ulothrix flaccida, when free from bacteria, were quite unable to live in a solution containing no nitrogen. Nostoc punctiforme could live perfectly well under these conditions provided that a drop of soil extract containing soil bacteria was added, and, moreover, the solution would show a decided increase in nitrogen content, showing that fixation of atmospheric nitrogen had taken place. The other two species, however, were incapable of living under similar conditions. Later, in 1897 and 1898, he showed that Nostoc punctiforme could live in association with soil bacteria in the absence of combined nitrogen, and also in darkness, provided that glucose is supplied. Without glucose no growth takes place in darkness. A very interesting experiment was recorded by the same worker in 1903, in which he showed that algae and soil bacteria, inoculated together into sterile sand with mineral nutrients devoid of combined nitrogen, are capable of fixing enough atmospheric nitrogen to support the growth of higher plants.

The idea of a symbiotic relationship between algae and soil bacteria has since gradually grown in popularity. Reinke ('03b) believes that *Volvox* colonies are ordinarily infested with the nitrogen-fixing organism *Azotobacter*, that the bacteria obtain some carbohydrate food from the gelatinous matrix of the colony,

and in return give up some combined nitrogen, in which they are very rich. In a somewhat earlier paper the same author (Reinke, '03a) extends this idea to marine algae, and expresses the probability that here also the algae are dependent upon Azotobacter for their nitrogen supply, and that the bacteria live embedded in the gelatinous surface of the algae.

Fischer ('04) asserts that there is a similar symbiosis between a terrestrial species of Oscillatoria and Azotobacter, and that even when the bacteria are not readily visible they can be made to increase rapidly by culture in 1 per cent mannite solution. It is interesting to note that he was unable to obtain the bacteria from Hormidium parietinum and Pleurococcus vulgaris inhabiting the bark of trees.

In opposition to all the work enumerated above and many other similar investigations too numerous to mention, all dealing with impure cultures or with algae and bacteria in mixed culture, indicating an increase in nitrogen content of the culture medium which was sometimes erroneously attributed to the algae themselves, is a group of articles dealing with experiments on algae in pure culture, free from bacteria, which give more conclusive evidence of the relation of algae to atmospheric nitrogen.

The earliest of these workers with pure cultures was Kossowitsch ('94) who isolated an alga from soil and proved quantitatively that no fixation of nitrogen could be demonstrated in the case of the pure alga, but that if impure cultures containing bacteria are used considerable fixation takes place. Kossowitsch realized the possibility of a symbiotic relationship.

Krüger and Schneidewind ('00) carried out very extensive experiments from which they drew similar conclusions. Working with about 8 species of *Stichococcus*, as many of *Chlorella*, and about 6 species of *Chlorothecium*, they proved that not one of these is able to live in a solution containing no combined nitrogen, and that even when growing vigorously there is no fixation of atmospheric nitrogen. They attribute the increased fixation in mixed cultures to the beneficial action of the symbiotic relation between the algae and bacteria rather than to the activity of the algae themselves.

Charpentier ('03), working with *Cystococcus humicola*,¹ also came to the conclusion that in pure culture this species is incapable of fixing atmospheric nitrogen.

Heinze ('06), working with a species of *Nostoc*, claims to have proved that fixation of nitrogen occurred in this species, but his alga was slightly infected with a species of *Streptothrix*. After isolating the fungus Heinze proved that this, by itself, was unable to fix atmospheric nitrogen, whereas the alga with its slight infestation showed a decided fixation. Heinze thus considered it proven that the fungus played no part in the nitrogen fixation, which he thought was due entirely to the alga. Since this worker confesses that he was unable to free his alga from the fungus, it is possible that bacteria were also present in the culture.

Schramm ('14), who gives a very complete survey of all the earlier work on the subject, working with 7 diverse species of soil algae in pure culture, proved that when no combined nitrogen was provided, not one of these algae was able to grow. This seems to indicate that they are incapable of making use of atmospheric nitrogen.

Nakano ('17), in the course of his paper, gives a more thorough investigation of the symbiosis between Azotobacter and algae than any other worker. He used species of Chlorella, Scenedesmus, etc., in pure culture, and also isolated and named from soil a pure strain of Azotobacter. Using a solution containing no combined nitrogen and .5 per cent glucose, Nakano found that the algae were unable to produce any growth, and moreover fixed practically no nitrogen when grown in pure culture in such a solution. Azotobacter, however, grew well under the same conditions and fixed atmospheric nitrogen abundantly. Mixing the pure Azotobacter and the pure algae in the same culture solution, a good growth of algae resulted, and in addition the amount of nitrogen fixed by the mixed culture of Azotobacter and algae was 20 per cent higher than by the pure strain of Azotobacter alone. This clearly proves the beneficial effect of the association of the two organisms. In further discussion concerning the more exact nature of the symbiosis, Nakano comes to the conclusion that the algae live at the expense of nitrogenous compounds derived from

<sup>&</sup>lt;sup>1</sup> Probably identical with Chlorococcum humicola (Näg.) Rabenh.

the Azotobacter, which they obtain after the death and autolysis of the bacterial cells. A sterilized culture of Azotobacter is not capable of furnishing the nitrogen necessary for algal growth. since the nitrogen is unavailable in a complex organic food, and enzyme action is destroyed. Debating on the conditions in nature, Nakano is inclined to believe that the symbiosis is not of any real importance to the algae, for he argues that whereas many thousands of Azotobacter cells are necessary to supply nitrogen for a single algal cell, one never sees algae thus infested in nature, and he is very doubtful whether Azotobacter could supply the nitrogen required by the larger seaweeds. In explaining the beneficial action of the algae on the nitrogen-fixing capacity of Azotobacter, Nakano thinks that possibly the oxygen evolved by the algae during photosynthesis is responsible for this. Azotobacter is more active if grown in thin layers than in thick layers and thus it seems to have a high oxygen requirement. The presence of algae might therefore be helpful so long as photosynthesis is going on.

In the past few years very few workers have claimed that algae by themselves are capable of fixing atmospheric nitrogen. Benjamin Moore and Webster ('20), and later Benjamin Moore, Whitley, and Webster ('21) have performed certain experiments and claim that nitrogen fixation is possible in both freshwater and marine algae. They made no attempt, however, to free their cultures from bacteria, and moreover they were apparently working in complete ignorance of the extensive literature on the subject and the results of the many workers who had preceded them. They were apparently also unaware of the fact, long proven at that time, that a symbiotic relationship exists between nitrogen-fixing bacteria and algae. Their results cannot therefore be considered to add anything to our knowledge of the relation of green algae to atmospheric nitrogen, although their philosophic discussions are interesting.

Wann ('21), on the other hand, used only pure cultures, and he also claims that nitrogen fixation is possible under certain conditions in green algae. Using 7 different species, he supplied his algae with nitrogen in several forms, in organic combination or as ammonium or calcium salts, in series with and without glucose.

Analysis was made of the media before the experiment, and of the culture solutions at the conclusion, by the Gunning-Kjeldahl method, and it was found that where the nitrogen was provided in the form of nitrate and in the presence of glucose, there was an increase in nitrogen content amounting to as much as 54 per cent. The author therefore concludes that the algae had fixed atmospheric nitrogen to that amount. Wann's work occupies a very isolated position, being the only case in which fixation of atmospheric nitrogen by green algae in pure culture has been claimed.

Muenscher ('23), working on the nitrogen metabolism in *Chlorella*, states incidentally that his results do not conform with those of Wann and that he always recovered the same amount of nitrogen after the experiment as was provided at the beginning. He was unable to demonstrate nitrogen fixation in this species.

Wann's work has been criticized by Bristol and Page ('23). These workers have carefully repeated Wann's experiments, and find that there is no fixation of atmospheric nitrogen by the algae used by them. They claim to have found the weak point in Wann's work to be the method of analysis when nitrates are present. They seem to prove their point fairly conclusively, and as far as one can judge from the evidence at present it seems quite likely that Wann's results will have to be held in abeyance until an explanation can be given of why his results are at variance with the work of other investigators.

Summarizing our knowledge of the relation of algae to nitrogen, it seems to be fairly well established that green algae in pure culture, free from bacteria, are unable to fix atmospheric nitrogen, but that when cultivated in association with Azotobacter, their presence seems to be beneficial, so that the capacity of the bacteria for fixing nitrogen is stimulated. The relation between the two organisms is probably of a symbiotic nature, the bacteria deriving carbohydrate food material from the gelatinous sheaths of the algae, and the algae thriving on the nitrogenous material provided by the bacteria.

# THE RELATION TO LIGHT AND CARBON

In the consideration of the peculiar conditions in which algae must live if they are present beneath the surface of the soil in an active condition, their relation to darkness and capacity for saprophytic nutrition must be important. According to Bristol, the number of algae in the soil at 4 inches below the surface is nearly as great as in the surface inch. Furthermore, the quantity is far greater than usually supposed. "Taking 100,000 as a rough estimate of the number of algae per gram of manured soil in a given sample and assuming the cells to be spherical and of average diameter,  $10~\mu$ , it has been calculated that the volume of algal protoplasm present was at least three times that of the bacteria, though only one-third that of the protozoa." [Russell, The Micro-Organic Population of the Soil, p. 110. 1923.]

Such great numbers of algae living at the depths at which they are found must be in complete darkness and under conditions which render photosynthesis impossible. A considerable amount of work has been done on the physiology of the algae, and the question of their capacity for saprophytic nutrition in the absence of light has received ample attention. There is so much literature on the subject that only a few of the cases in which saprophytic nutrition has been demonstrated can be mentioned.

Radais ('00), working on *Chlorella vulgaris*, found that his cultures remained green in darkness and also that when grown on potato malt agar, growth was equally good in light and in darkness, while Grintzesco ('03), working with the same organism, obtained exactly the same results, and even states that with 2 per cent glucose, growth in total darkness may be much better than in the light. Artari ('06) and Kufferath ('13) also support these statements.

Charpentier ('02, '03) found that Cystococcus humicola¹ grew and remained green in darkness, but that the yield was much greater in the light, a weight of 330 mgms. resulting in the light as against 27 mgms. in darkness. Artari ('02) also found that the same alga produced normal healthy growths if grown in darkness with 1 per cent mannite, lactose, glucose, levulose, or canesugar.

Treboux ('05) has shown that other substances than carbohydrates may be used by algae as a source of carbon in the absence of light, for he obtained growth with several species of algae in

<sup>&</sup>lt;sup>1</sup> Probably Chlorococcum humicola (Näg.) Rabenh.

darkness, using the potassium salts of various organic acids and also the amino derivatives, such as glycocoll, alanin, leucin, and asparagin. With the acids, the simplest compounds seemed to be the most easily assimilated, and a concentration of .25 per cent acetic acid was used by practically all the 40 species of algae investigated.

Chlamydomonas Ehrenbergii, according to Artari ('14), is able to use glucose in darkness, for if glucose is present the yield in the absence of light is 20–25 times greater than if no sugar is given. However, the growth is at no time so good as when autotrophic nutrition is permitted.

Dangeard ('21) records that he has grown Scenedesmus actatus in darkness for 8 years. One per cent glucose and .8 per cent peptone were provided, and frequent transfers to new media were made. After being subjected to darkness and prevented from exercising its photosynthetic function for so long, exposure to light resulted in the evolution of oxygen in 5 hours. This is very interesting in connection with the soil algae question.

The above work, confined to species of the Chlorophyceae, is altogether in favor of the possibility of heterotrophic nutrition in darkness. Not so much work of this kind has been done on the Cyanophyceae, and the evidence is not nearly as conclusive. Bouilhac ('97) found that Nostoc punctiforme would grow in darkness provided that glucose was present. His experiment, however, is complicated by the fact that he was using a mixed culture with soil bacteria. Pringsheim ('13) isolated several species of Nostoc and Oscillatoria, but found that the addition of organic matter to the cultures produced deleterious effects if used in large quantities, and that the stimulating action of small amounts was never very striking. He was unable to demonstrate that saprophytic nutrition with organic food was possible in the dark.

# CULTURAL METHODS AND RESULTS

The culture vessels were prepared in essentially the same way as described in an earlier paper (Moore and Karrer, '19) except that the sand was well washed before being used and the culture solution was full strength. It was not felt necessary to slant the

bottles, since the sides of the bottle served as a suitable substratum for forms not requiring aquatic conditions. All the samples were taken from excavations in the Missouri Botanical Garden in localities where the soil had not been disturbed for at least twenty-five years, so that the results obtained should be regarded as pertaining to uncultivated rather than to cultivated soils. For the most part the several localities had been covered with a The depth from which the samples were taken was noted at the time, and every precaution observed to prevent contamination of the samples with soil from a different level. case of the first 3 series of cultures the culture vessels were inoculated in the garden with an unknown amount of soil, but in the fourth series the samples were taken in sterile bottles, and inoculation was performed in the laboratory with weighed amounts of In order to prove that infection from the air did not take place during the weighing of the inoculum, a sterile control culture was left exposed to the air of the laboratory for 36 hours during the weighing of the soil, but no growth of algae resulted from this exposure. The algae developing in the cultures can therefore be assumed to have developed from the soil with which they were inoculated.

The first 3 series of cultures were inoculated with soil of different levels down to 4 or 5 feet, but in the last series the samples were taken as deep as 9-10 feet.

The results recorded here can be considered as amplifying the data given in the earlier paper (Moore and Karrer, '19), not only in extending our knowledge of the depth at which algae occur in the soil but also in giving an idea of the variety of the subterranean flora. Although by no means as luxuriant as at the surface (as investigated by Bristol, '19a, '21), the flora nevertheless includes a far greater number of species, especially *Chlorophyceae*, than one would expect in subterranean conditions.

The species which is almost universally present is *Chlorococcum humicola*, recorded as *Protoderma viride* in the earlier paper. It should be noted that Bristol ('19b, '21) has obtained this alga from the Malay States, and has also found it usually present in English soils. It therefore seems likely that this species is universally present in all soils. In the present cultures it appeared in almost

every case where a rough inoculum (about 10 gms.) of soil down to 3 feet was used, and it will probably occur in samples as small as .1-.2 gms.<sup>1</sup> Below 3 feet, however, its occurrence is so uncertain that one cannot be sure of obtaining it unless more than 5 gms. of inoculum are used.

Chlorococcum humicola is accompanied by a number of other green algae, which seem to inhabit the soil in somewhat smaller numbers than this dominant species, so that some, but not all, may occur along with Chlorococcum in most of the cultures. Below 3 feet it not infrequently happens that one or other of these species may be obtained in a unialgal culture, a circumstance which aided considerably in their identification as distinct species.

Whereas Chlorococcum humicola occurs with sufficient constancy for it to be possible for us to come to some conclusions regarding its numerical distribution in the lower layers of the soil, most of the other species seem to be very uneven in their distribution and to occur without any regularity, so that it is impossible to make any definite statements concerning their numbers in the soil.

It is noteworthy that these accessory species do not correspond very closely to the list of *Chlorophyceae* enumerated by Bristol ('21) as accompanying *Chlorococcum humicola* in the English soils. One species which is given by her, namely, *Chlorochytrium paradoxum*, is, however, more constantly present in the Missouri samples than in England. Apart from this, there is almost no conformity between the two lists, and one is forced to the conclusion that the subterranean flora is probably as variable as the surface flora.

The most interesting species isolated from the Missouri soils and not present in Miss Bristol's list is *Protosiphon botryoides*, nearly always present in our cultures (and possibly "Cladophora sp." in Moore and Karrer, '19). This species is not native in England and is therefore not likely to occur in the subaerial flora. The frequent occurrence of Chlorella in our cultures, and its absence from the British list is, on the other hand, somewhat surprising, and one cannot help thinking that possibly Miss Bristol

<sup>&</sup>lt;sup>1</sup> The weight of inoculum must only be regarded as approximate, since the moisture, which varied in samples from different levels, was not taken into account.

overlooked it, or mistook it for small cells of Chlorococcum humi-cola.

Botrydiopsis arhiza is another species which is probably more often present than is apparent from the accompanying lists, since isolated cells are easily overlooked. As regards the species of Chlamydomonas, it is possible that Miss Bristol has included at least one species in her stages in the life history of Chlorococcum humicola ('19b). The form figured by her in pl. 18, figs. 27, 28, is very similar to the alga recorded here as Species B. Chlorococcum humicola has been isolated by one of us in pure culture, and a palmelloid stage has never been observed to occur. Species B has also been isolated, and it always retains its characters both on solid and liquid media.

Species of Cyanophyceae and diatoms are notably much fewer than recorded in Miss Bristol's surface flora, and the Cyanophyceae only occurred in dominating numbers as a rule in the upper 18 inches of soil. On the other hand, the development of Lyngbya subtilis, 2½ feet down, in Series A, is suggestive of the possibility that in the method of culture used the development of Chlorophyceae was perhaps unduly favored. The obtaining of a pure culture of Oscillatoria amphibia from a depth of 8 feet 2 inches in Series D is likewise a very interesting fact, which was considered at first as being due to a chance contamination from a different level. However, since the greatest precautions were taken during the collecting of the samples and the inoculation of the cultures, and, moreover, since isolated cultures of different species were obtained from samples of soil taken from 5 to 9 feet in about 8 different cultures, it seems unlikely that foreign infection was responsible for all 8 cultures. This is especially true since with one or two exceptions the constituents in these cultures are not at all common in the cultures from higher levels, and represent in some cases the only record of that particular species. If the development of these cultures from lower levels is due to infection with soil from a higher level, the species most commonly occurring there, namely, Chlorococcum humicola, would be expected to appear.

The diatoms are not fully recorded here for the first 18 inches, since a few other species sometimes occurred in the uppermost

samples taken, but in too small numbers for the cleaning up of a sample for specific determination to be a successful operation. The records for the lower depths can be considered fairly accurate. The record of *Navicula atemoides* in pure culture at a depth of 5 feet 5 inches is extremely interesting, and this is the species most constantly present in cultures from the lower strata.

In identifying the species the greatest difficulty was experienced in deciding between *Chlorochytrium paradoxum* and *Protosiphon botryoides*, and it was often impossible to determine whether one or both of the species were present. *Protosiphon*, after a length of time in culture, proceeds to form large resting aplanospores which may be released from the old mother cell-wall and remain for a long period as orange-colored cysts. *Chlorochytrium paradoxum* also forms similar large cysts which cannot be distinguished from those of *Protosiphon*. The records for these two species are therefore somewhat uncertain.

The succession of species in the cultures was very interesting, for with age, species originally dominating would fade away and give place to others. *Protosiphon* begins to lose its typical form after about 6 months' culture, probably because an aquatic habitat is not normal for it. The most interesting case, however, is that of the alga recorded as Species A, which was never observed until a culture reached the age of 8–12 months, except in the culture 42C where it occurred as a unialgal culture and formed a recognizable but feeble growth in 3 months. Probably high organic food requirement is responsible for these facts.

It should be noted that series D produced a large number of species which were not observed at all in the three earlier series. Possibly this is due to the fact that a greater proportion of unialgal cultures occurred in this series. These rarer species may have been present in the other series, but were not conspicuous because of the greater mass of other forms, or again the difference may be due to a local variation in the flora.

## NOTES ON THE SPECIES OBSERVED

#### CYANOPHYCEAE

#### Nostoc commune Vauch.

Forti in De Toni, Syll. Alg. 5: 404. 1907.

This species occurred in a well-developed form in several cultures, all above 1 foot 4 inches. The trichomes were about 5  $\mu$  in diameter, with heterocysts very slightly larger than, or equal in size to, the vegetative cells. Spores were abundant in long chains and measured about  $6\times 8\,\mu$ .

## Nostoc muscorum Ag.

Forti in De Toni, Syll. Alg. 5: 400. 1907.

This species was identified in only one culture, from a depth of 6 inches. The spores were somewhat immature, but the other characters seemed sufficient to identify it.

#### Nostoc comminutum Kütz.

Forti in De Toni, Syll. Alg. 5: 393. 1907.

A pure culture of an alga corresponding to this species was obtained from soil at a depth of 3 feet 6 inches. The colonies were small and distinct from each other, consisting of trichomes 3–3.5  $\mu$  in diameter, closely convoluted. Heterocysts 6.5  $\mu$  in diameter were observed, but spores were wanting. The occurrence of a blue-green alga at such a depth is somewhat surprising.

# Phormidium tenue (Menegh.) Gom.

Forti in De Toni, Syll. Alg. 5: 227. 1907.

This species was identified in only one culture, at a depth of 1 foot. It was quite typical.

# Phormidium molle (Kütz.) Gom.

Forti in De Toni, Syll. Alg. 5: 219. 1907.

In the same culture as the preceding species was a small quantity of a very moniliform *Phormidium* with trichomes  $3.5 \,\mu$  in diameter, which seemed to correspond fairly well with the description given for this species.

# Lyngbya subtilis West

Forti in De Toni, Syll. Alg. 5: 285. 1907.

In a culture of soil from a depth of  $2\frac{1}{2}$  feet there developed after a time a quantity of a very slender blue-green alga with a distinct sheath. The filaments were about  $1.5\,\mu$  in diameter, and the trichomes themselves barely  $1\,\mu$ . The alga seemed nearest to Lyngbya subtilis West.

# Oscillatoria amphibia Ag.

Forti in De Toni, Syll. Alg. 5: 169. 1907.

The development of a pure culture of this species from soil of more than 8 feet depth was a very surprising phenomenon. The alga was in every way normal and typical.

#### BACILLARIEAE

#### Navicula atemoides Grun.

Van Heurck, Diat. 227. pl. 5, f. 230. 1899.

This was the most general of all diatoms, and was a general constituent of the flora from the surface 6 inches to a depth of 5 feet 5 inches, which is the lowest record obtained for it.

# Navicula mutica Kütz.

Van Heurck, Diat. 206. pl. 4, f. 167. 1899.

This species was not quite as frequent as the preceding one, but nevertheless has been found at a depth of 4 feet.

# Hantzschia amphioxys (Ehr.) Grun.

Van Heurck, Diat. 381. pl. 15, f. 483b. 1899.

This is one of the less frequent diatoms.

# Nitzschia palea (Kütz.) W. Sm.

Van Heurck, Diat. 401. pl. 17, f. 554. 1899.

Rather more frequent than the preceding species and observed below 4 feet.

#### CHLOROPHYCEAE

# Chlorococcum humicola (Näg.) Rab.

Bristol in Jour. Linn. Soc. Bot. 44: 473. 1919.

This alga developed in nearly every culture down to 3 feet, and the lowest record is 5 feet 5 inches. As will be seen from table 1v, series D, .01 gm. of soil at 12 inches depth gave a good growth of Chlorococcum. It can therefore be assumed that at this depth there are at least 100 individuals of the alga to the gram. At 3 feet depth, however, they may be as sparse as one individual in 2 gms., and at 4 feet 2 inches rarer than one in 5 gms. The alga reproduced freely by both zoogonidia and aplanospores, and the vegetative cells readily formed orange resting cysts when conditions became unfavorable for vegetative growth. There is some doubt as to whether the large cells mentioned by Bristol ('19b) really belong to this species. They possibly belong to Chlorochytrium. Furthermore, the palmelloid stage mentioned and figured by Bristol probably belongs to a species of Chlamydomonas, Species B of this work (vide supra).

# Chlorochytrium paradoxum (Klebs) West

Bristol in Jour. Linn. Soc. Bot. 45: 8. 1920.

This species was a fairly frequent constituent, though it was often difficult to decide for certain reasons whether it was really present or not, especially when *Protosiphon botryoides* was also present (vide supra). It usually occurred in the form of large olive-green cells or bright orange resting cysts, reaching a diameter of  $100~\mu$ . There were rarely any of the smaller cells showing the characteristic cytological structure of the genus, and the only way in which the identity of the cysts was suspected was in following the development of the zoogonidia into the characteristic vegetative cells. Large aplanosporangia with a few comparatively large cysts of unequal size were sometimes observed, all in a resting condition. The thickness of the wall of the aplanosporangium might be as much as  $7{\text -}10~\mu$ .

# Protosiphon botryoides (Kütz.) Klebs

Klebs, Bed. Fortpflanz. 169-222. pl. 1, f. 1-16. 1896.

This species was a very frequent constituent of the cultures, being present in practically all samples in series B and C. Very often it occurred in great abundance, so that it was probably present in considerable quantity in the soil. In the cultures the alga was not always typical in form because of the aquatic conditions but it was always easily recognized. Reproduction by aplanospores of varying size was frequent, and the production of gametes, their conjugation, and the formation of the tiny starlike zygotes figured by Klebs (loc. cit., pl. 1, f. 16) were also observed. Sometimes the aplanospores were transformed into large, orange, thick-walled resting cysts. The lowest depth from which the species has been obtained is 4 feet.

# Chlorella spp.

The genus Chlorella was represented fairly constantly in the cultures, and it seems quite possible that more than one form occurs. The specimens isolated from a certain culture measured uniformly 3–5  $\mu$  in diameter, whereas in other cultures they were only 2–3  $\mu$  in diameter. This indicates that possibly two forms occur in the culture, but in the records no distinction is made between the two. Chlorella has been observed as far down as 5 feet.

# Trochiscia reticularis (Reinsch) Hansg.

West, Brit. Freshw. Alg. f. 82K. 1904.

This species occurred in 3 of the 4 series of cultures, and in the last series, D, was almost as constant and as abundant a constituent as Chlorococcum humicola itself, the indications being that there are more than 100 specimens per gram at a depth of 12 inches. It must be noted, however, that whereas .01 gm. of soil from this level produced individuals of Trochiscia, 5 gms. from the same level failed to produce any specimens. The distribution of this species therefore seems to be very uneven. The specimens varied in size from 10 to 20  $\mu$  in diameter, and were sometimes oblong rather than spherical in outline, with the dimensions 12  $\times$  8  $\mu$ . The lowest record for the species is 3 feet 9 inches.

# Trochiscia sp.1

? Acanthococcus sp., Reinsch, Ber. d. deut. bot. Ges. 4: 243. pl. 12, f. 18. 1886.

This species was observed only in a single culture, at a depth of 3 feet 9 inches. It was sharply distinguished from *Trochiscia reticularis*, which was the more common species of the genus, by its short, blunt, and rounded projections. The specimens varied in size from 14 to 18  $\mu$  in diameter and seemed nearest to the form recorded by Reinsch as *Acanthococcus* sp.

## Dactylococcus sp.

Cells of this form were observed only in two cultures. They were stout and spindle-shaped with slightly acute apices, and measured about  $12 \times 8 \,\mu$ . A pyrenoid was distinctly present, and reproduction was observed by the formation of 4 similar individuals within a mother cell, which might at this time reach the size of  $12 \times 16 \,\mu$ . In general appearance the form was most suggestive of the *Dactylococcus* state of *Scenedesmus* figured by Grintzesco ('03, p. 217), but what it really is was not decided. Bristol ('21, p. 74) records a species of *Dactylococcus* from English soils, but there can be no confusion between the form observed here and her species.

#### Ulothrix subtilis Kütz.

West, Brit. Freshw. Alg. 76. f. 20 C-F. 1904.

This species is not at all a frequent constituent of the subterranean flora, although, according to Bristol ('21), it is almost universally present on the surface. In two of the series of cultures it did not occur at all, and its occurrence in the other two series was sporadic. The record at 3 feet in series D is somewhat surprising. It seems probable that for some reason this alga does not usually descend very far from the surface of the soil, possibly owing to the fact that its zoogonidia are not long motile. The filaments were  $5-7.5~\mu$  in diameter, and the cells almost as long as, or a little longer than, broad. There was some tendency for the filaments to break up into short lengths.

<sup>&</sup>lt;sup>1</sup> Bristol ('21) records two species of *Trochiscia* from English soils, but neither seems to be identical with the forms observed here.

# Stichococcus bacillaris Näg.

West, Brit. Freshw. Alg. 80. f. 24A. 1904

This species does not seem to occur with any regularity, unless, as is quite possible, the tiny cells were often overlooked. It was observed in only 3 cultures, the lowest record being 2 feet 6 inches.

# Stichococcus scopulinus Hazen

Hazen in Mem. Torr. Bot. Club 11: 161. pl. 22, f. 4-6. 1902.

This alga was observed in only two of the series and never below a depth of 12 inches. The cells were 3–4  $\mu$  in diameter and 11–16  $\mu$  in length. The filaments were often of considerable length and showed little tendency to dissociate. It seems likely that the occurrence of this species in the subterranean flora is dependent upon its local distribution at the surface and that it probably never descends to any great depth.

#### Uronema confervicola Hazen

Collins, Green Alg. N. Am. 88. f. 66. 1909.

The occurrence of a few isolated filaments of this species in 2 cultures from a depth of 3 feet was indeed surprising. The filaments were about  $4 \mu$  in diameter and of great length. The cells were provided with 2 pyrenoids each, and the apical cell was typically acute. The species is probably not a regular inhabitant of the soil but only a chance form. In both cultures it was present in such small quantity that a month after it was first observed it had quite disappeared.

#### Monocilia viridis Gerneck

Gerneck, Beih. Bot. Centralbl. II. 21: 263. pl. 12, f. 77-84. 1907.

A form closely resembling this alga described by Gerneck in structure and appearance was observed in 3 cultures of series A. In the culture from the lower level,  $3\frac{1}{2}$  feet, it was present in considerable quantity and for a time favored the dominant constituent of the culture. Later it disappeared entirely from both cultures and left no trace. In the culture in which it had been most abundant, a considerable quantity of a yellow-green alga

appeared which was identified as Botrydiopsis arhiza Borzi. In this connection it is interesting to note that Gerneck states that Monocilia viridis, after being cultured for about 2 months in a liquid medium, loses its branched and filamentous form and goes into a unicellular palmelloid state. According to Gerneck, the filamentous form can only be obtained from the palmelloid stage by cultivating on a solid medium such as agar. The alga identified as Botrydiopsis arhiza in these cultures has been grown on agar, however, and it always retained its unicellular form. If Gerneck's observations are correct, therefore, it would seem that Monocilia viridis and Botrydiopsis arhiza as observed in these cultures are distinct from each other. It may be that Monocilia viridis is a more constant inhabitant of the soil than would appear from these records, but that it is not often in a recognizable condition.

# Botrydiopsis arhiza Borzi

Borzi, Studi Alg. 2: 169. pl. 12, 13. 1895.

This is the most regular representative of the *Heterokontae* in the subterranean flora. It is rarely present, however, in great abundance, and is possibly often overlooked. When there is not too much competition with other species it multiplies rapidly, however, and may form an abundant growth. Reproduction by aplanospores was very common, and zoogonidia with only one visible cilium were also observed. On one occasion biciliate swarmspores, similar to the gametes figured by Borzi, were seen, but no conjugation took place. The lowest depth from which the alga was obtained was 4 feet.

# Characiopsis minuta Borzi

Borzi, Studi Alg. 2: 152. pl. 14, f. 1-12. 1895.

The occurrence of this species in a single culture was somewhat surprising. It was present in considerable quantity in a sample taken from a depth of 2 feet 3 inches and only differed from the typical form in its slightly smaller size. The finding of this species seems to indicate that the spores of many algae may occasionally find their way into the soil and suggests that the subterranean flora may prove, with increased investigation, to be

almost as rich in species as the surface flora, though not in numbers of individuals.

# Species A

The form recorded under this heading is a very problematical one which, since swarmspores have not yet been observed, cannot be properly identified. It is probably a fairly constant inhabitant of soil, but evidently requires very special conditions for its development, for it only appears in old cultures. The cells float freely in the water, quite isolated from each other and without any tendency to adhere in colonies. They are usually oval-oblong,  $10-28~\mu$  long by  $7-18~\mu$  broad, though quite frequently they may be spherical with a diameter of  $9-15~\mu$ . In some instances a number of unusually large individuals may occur scattered among the smaller ones, either spherical or oval, and reaching a diameter of  $40~\mu$ .

The most striking feature of the alga is the presence of a bright red spot in the interior of the cell. This is obviously not a stigma, for it is much larger, reaching a diameter of 2–8  $\mu$ . The chloroplast is a small parietal plate which only covers part of the wall. There is neither a pyrenoid nor starch present, though oil is abundant. The systematic position of the alga is unknown.

# Species B

This is most probably a species of Chlamydomonas, and was the most frequently encountered representative of the genus. It is in all probability more constantly present than is indicated by the tables, and, especially in the earliest examination of series A, was possibly very often mistaken for stages of Chlorococcum humicola. Complete isolation of the form has shown, however, that it is a distinct species. It usually occurred as oval cells embedded in a gelatinous stratum, and if present in any great quantity, or if pure, forms a soft gelatinous stratum similar to masses of Tetraspora. A slight change in temperature causes the green cells to acquire cilia and to swim out of the gelatinous matrix. The period of swarming is comparatively short, and the cells soon become quiescent again and secrete quantities of mucilage to form a large expanded colony as before. Multiplication takes place in the palmelloid stage by the division of individual cells,

usually into fours. The motile cells are oval in form, 7–12  $\mu$  long by 3–7.5  $\mu$  wide. They have a massive bell-shaped chromatophore which only leaves a minute rounded clear space at the apical end of the organism. There is a distinct pyrenoid, usually in the posterior region, but sometimes more or less lateral. The stigma is distinct and somewhat elongated, lying in the apical region of the cell. The two cilia are about equal to the body of the organism in length. Bristol ('21) does not record the occurrence of this form, but figs. 27, 28 of pl. 18, in her work on Chlorococcum humicola ('19b), are identical in appearance with the forms observed in the present cultures, and possibly represent the same organism. It is noteworthy that this species has the record for depth, having been obtained at a depth of more than 9 feet.

# Species C

This form is apparently another species of Chlamydomonas. It was only observed with certainty in a single culture in which it occurred pure. It may perhaps have been present in other instances, being possibly overlooked in the confusing mixture of The macroscopic appearance of the culture is very different from that of the preceding species, for the alga, instead of forming a large gelatinous stratum, as in Species B, produces small tough green flakes. Microscopically, the cells are somewhat rounded, possess a distinct pyrenoid, and are arranged in gelatinous clumps of greater or smaller size, though never forming such a large expansion, or possessing so much mucilaginous material as Species B. A slight change in the external conditions induces the development of cilia, as before, and the cells become motile and swim out of the gelatinous stratum. The motile cell is a little stouter than in Species B, reaching a length of 5-9 u and a breadth of 3.5-6 μ. The chloroplast is not so distinctly bellshaped; it leaves a larger and more irregularly shaped space clear at the anterior end, and the stigma is very minute and difficult to find, and also, when visible, is more anterior in position. The chief differences between the two species are to be found in the chloroplast and stigma and in the macroscopic nature of the colonies. It is possible that the Protococcus-like stages referred to above may belong to this species.

## Species D

In many of the cultures gelatinous masses were frequently observed among the other forms, in which the cells embedded in the gelatinous stratum possessed the stigma and other characters of The slightly changed conditions resulting from the removal of the sample from the large culture vessel, and its examination on a slide almost invariably induced the small cells to become motile. Then, in the earlier examination of the cultures. these motile cells were always observed to unite in pairs, producing An attempt was made, but without success, a rounded zygote. to follow the development of the zygote, and unfortunately the alga never occurred alone, or in sufficient quantity to make its isolation possible. In older cultures a similar form was frequently observed which agreed in size and in its conspicuous stigma, but the contents were so obscured by the presence of large starch grains that other cytological comparisons were impossible. Although in these older cultures the cells readily became motile as before, conjugation was not observed to occur.

The motile cells of this alga are smaller and more rounded than in the two preceding forms. They are  $3\frac{1}{2}-5~\mu$  in diameter, and only slightly longer than broad. The chloroplast is not particularly massive, covering only a part of the external wall, and contains a pyrenoid which is usually quite conspicuous. The stigma is distinctly visible and the cilia are somewhat longer than the body length. The form has been observed as far down as 4 feet.

# Species E

This is an additional species of Chlamydomonas which was not nearly such a constant constituent of the soil as some of the others, or at least it never occurred in quantities large enough to be conspicuous, although it may sometimes have been present as isolated individuals. It was never observed in a motile condition, but that it is normally a motile organism and probably a species of Chlamydomonas seem to be undoubted facts. It was readily distinguished from all other similar forms by its size, reaching a length of  $12-21~\mu$  and a breadth of  $10-12~\mu$ . The cells were usually broadly oval and occurred in most cases in the palmelloid form, each cell surrounded by its own distinct gelatinous envelope

which might reach a thickness of  $10\,\mu$ , and aggregated in larger or smaller clumps. Very often active cell division seemed to have been taking place, for 2–8 smaller individuals were sometimes seen crowded together in the same envelope. In the majority of the larger individuals it could be clearly recognized that the alga was normally a ciliated organism by the differentiation of the cell contents, a clear apical region being easily distinguished. Apart from this, too much reserve food, both starch and oil, was usually present for the cytological structure to be clearly seen. The lowest depth at which it was found is 4 feet.

# Species F

There was some doubt at first whether this organism is really an alga or a large bacterium, but the balance of evidence seems to be in favor of its being an alga, most probably of the genus Stichococcus. The cells are very minute, oblong and angular,  $1\frac{1}{2}-2$  u broad and 4-5 u long. They are distinctly, though faintly. green in color and seemed at first to have homogeneous contents. The higher magnification of the oil immersion showed, however, that in some individuals a clear space could be recognized either at one or both ends or else along the lateral margin. This seems to indicate that there is a chloroplast in the form of an extensive parietal plate. There is no pyrenoid, and very little blackening with the addition of iodine. The bacillus-like form of the organism at once distinguishes it from Stichococcus bacillaris. formation concerning its reproduction is to be desired before its exact affinities can be decided. The species was observed only in one or two isolated cultures.

# Species G

This is one of the several forms peculiar to series D, in a number of samples of which it occurred, including some from a depth of more than 6 feet. The cells are isolated and spherical, 9–13  $\mu$  in diameter, reaching in exceptional cases 19  $\mu$ . There was usually a single bright green chloroplast, occasionally two, but neither pyrenoids nor starch were present, their place being taken by oil. Reproduction occurred by the formation of aplanospores which were produced in large numbers within a mother cell. These

small aplanospores gradually increase in size until they reach the dimensions of the ordinary vegetative cells. Until the swarm-spores of the alga have been obtained, its systematic position cannot be stated.

### Species H

In two cultures from a depth of 3 feet, brownish patches appeared between the sand and the glass at the bottom of the cultures, which were at first thought to be due to diatoms, but which proved, on examination, to consist of minute organisms probably of a flagellate nature. The tiny cells were 2.5–3  $\mu$  wide by 4.5–5  $\mu$  long, and had a bell-shaped brownish green chromatophore lining the greater part of the outer membrane. There was no pyrenoid. The organisms were not observed in the motile condition, and nothing is known of their cilia. There was always a very conspicuous projecting stigma, however, which indicates that they are normally motile.

#### Species I

This puzzling form occurred in only two cultures, one from a depth of 5 feet 8 inches and the other 8 feet 7 inches. It consists of small oval cells 4–7.5  $\mu$  long and 3–5  $\mu$  wide. There is a single parietal chloroplast which often does not cover the entire wall, and in which a conspicuous pyrenoid is embedded. Multiplication takes place by the formation of 2–16 aplanospores within a mother cell. Motile stages were not observed, and there is no tendency to the formation of gelatinous colonies.

# Species J

? Ankistrodesmus Pfitzeri (Schröder) West, Brit. Freshw. Alg. 224. f. 94 G, H. 1904.

In association with the preceding species in culture 62 at a depth of 5 feet 8 inches were conspicuous elongated cells. In form they seemed to be very similar to Ankistrodesmus Pfitzeri, but they were somewhat smaller and perhaps also a little broader in proportion. The cells were about 10  $\mu$  long and 3  $\mu$  wide, and there was a parietal chloroplast but no pyrenoid. A. Pfitzeri is usually stated to occur in gelatinous colonies, but in this culture the cells, although tending to adhere to each other and therefore

being surrounded by some gelatinous material, did not occur in well-defined colonies. Unfortunately, the species did not persist for very long in the culture but was gradually crowded out by Species I.

#### Moss Protonema

Moss Protonema was observed in only one culture, at a depth of 6 inches. It can therefore be assumed that normally the filaments do not descend to any great depth as do some algae, but are comparatively superficial in their growth.

TABLE I

(Series A)

SAMPLES TAKEN NOVEMBER 18, 1922, BETWEEN THE EXPERIMENTAL GREEN-HOUSE AND THE OLD RESIDENCE

Number of culture	Depth	Date of openingtion	Date of examination	Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Monocilia viridis	Stichococcus bacillaris	Stichococcus scopulinus	Protococcus-like colonies	Chlamydomonas (unidentified)	Species A	Species B	Species D	Species E	Species F	Nostoc muscorum	Nostoc commune	Nostoc sp.	Lyngbya subtilis	Phormidium sp.	Navicula atemoides	Navicula mutica	Hantzschia amphioxys
1/2	6 inches	Apr. May Nov. Apr.	1923 1923 1923 1924	×	1						×		?	?	?			?	×	×	×××		×	×××	×	
1a			1923 1923 1923 1924	_			×××								×											
1b	12 inches			-	×		××	×	×				? ? ?	×			×									
2a	18 inches	Feb. Mar. Apr. Apr.	1923 1923 1923 1924	××××									×													

# TABLE I (Continued)

Number of culture	Depth	Date of owningtion		Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Monocilia viridis	Stichococcus bacillaris	Stichococcus scopulinus	Protococcus-like colonies	Chlamydomonas (unidentified)	Species A	Species B	Species D	Species E	Species F	Nostoc muscorum	Nostoc commune	Nostoc sp.	Lyngbya subtilis	Phormidium sp.	Navicula atemoides	Navicula mutica	Hantzschia amphioxys
2b	18 inches	Mar. Apr. Nov. Apr.	1923 1923 1923 1924	××××	×			×					×		×											
3	2 feet	Apr. Nov. Apr.	1923 1923 1924	×××			×																			×
4	2 ft. 6 in.	Mar. Apr. Nov. Mar. Apr.	1923 1923 1923 1924 1924	×××××	××					?			×	×××		×	×					××				
5	3 feet	Mar. Apr. Nov. Apr.	1923 1923 1923 1924		××××		×									×								××××		
6	3 ft. 6 in.	Feb. Apr. Nov. Apr.	1923 1923 1923 1924	×	×		××	×	×			×	×			×										
7	4 feet	Feb. Apr. Nov. Apr.	1923 1923 1923 1924	×××	×	×××	×××	×					××												×××	
8	5 feet	Nov. Apr.	1923 1923				×																			

TABLE II
(Series B)

8AMPLES TAKEN APRIL 19, 1923, NORTHWEST OF CENTRAL LILY POND,
MAIN ENTRANCE

						AAI	N	EL	TF	IAS	VC1	E										===
Number of culture	Depth of sample	Date of examination	Chlorococum humicolo	Chlorochultium naradorum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Ulothrix subtilis	Characiopsis minuta	Chlamydomonas (unidentified)	Species A	Species B	Species D	Species E	Protococcus-like stage	Gloeocystis-like stage	Nostoc sp.	Navicula atemoides	Navicula mutica	Nitzschia palea	Hantzschia amphioxys
26a	1 ft. 6 in.	May 192 Jan. 192 Apr. 192	23 > 24 > 24 >	? >	×	××		××			×			×								
26b	1 ft. 6 in.	May 192 Jan. 192 Apr. 192	23 > 24 > 24 >	X X X	×			×			×											
29a	1 ft. 6 in.	May 192 Feb. 192 Apr. 192	23 > 24 > 24 >		×××		17						×		×	×			×			
<b>2</b> 9b	1 ft. 6 in.	May 192 Feb. 192 Apr. 192	23 > 24 > 24 >	?	×	×			×		×	××						×	×	×	×	
25a	2 ft. 3 in.	May 192 Jan. 192 Apr. 192	23 > 24 > 24 >	?		×					×								×			
<b>25</b> b	2 ft. 3 in.	May 192 Jan. 192 Apr. 192	23 > 24 > 24 >	\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\						×	×					×	×		×			
24a	2 ft. 6 in.	May 192 Nov. 192 Mar. 192 Apr. 192	23 > 23 > 24 >	?	××?×	××					×			×		×			××			×
24b	2 ft. 6 in.	May 192 Dec. 192 Apr. 192	231>		×						×			×					×××			
27a	2 ft. 6 in.	May 192 Jan. 192 Apr. 192	3 > 4 > 4 >	?	×××						×			×					×			
27b	2 ft. 6 in.	May 192 Jan. 192 Apr. 192	23 > 24 > 24 >	?	×××						×			×								

# TABLE II (Continued)

Number of culture	Depth of sample	Doto of own in this		Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Ulothrix subtilis	Characiopsis minuta	Chlamydomonas (unidentified)	Species A	Species B	Species D	Species E	Protococcus-like stage	Gloeocystis-like stage	Nostoc sp.	Navicula atemoides	Navicula mutica	Nitzschia palea	Hantzschia amphioxys
23a	3 feet	May Nov. Apr.	1923 1923 1924	×××	?	×××	×					×								×××			
23b	3 feet	May Nov. Apr.	1923 1923 1924	lΧ		×××	×					×								×			
28a	3 feet	Feb.	1923 1924 1924	×××	×	×	××					×			×					××			
28b	3 feet	May Feb. Apr.	1923 1924 1924	×××	?	×××	×					×		×				×		×××			
22a	3 ft. 6 in.	Nov.	1923 1923 1924	×××	?	×	×					×		×									
<b>22</b> b	3 ft. 6 in.	May Nov. Apr.	1923 1923 1924	×××	?	××	××					×		×		-							
21a	4 feet	May Nov. Apr.	1923 1923 1924	×××	?	×××	×					×					×						
<b>21</b> b	4 feet	May Nov. Apr.	1923 1923 1924	×××	X??	×××	×					×			×								
30	4 feet	May Feb. Apr.	1923 1924 1924	1 3		×	×	×				×				×		×		×			_

# TABLE III (Series C) SAMPLES TAKEN JUNE, 1923, SOUTH OF GRADUATE LABORATORY

															=					_	==		
Number of culture	Depth of sample	Doto of our minotion	Date of examination	Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Trochiscia sp.	Chlamydomonas (unidentified)	Dactylococcus sp.	Species A	Species B	Species D	Species E	Gloeocystis-like stage	Nostoc commune	Nostoc sp.	Phormidium tenue	Phormidium molle	Naricula atemoides	XX   Nitzschia palea
50	1 foot	Feb. Apr.	1924 1924	×			×					41/		×				×	×	×	×	×	×
49	1 ft. 4 in.	Oct. Feb. Apr.	1923 1924 1924	×××		××								×		×		×				×	
48a	1 ft. 8 in.	Oct. Feb. Apr.	1923 1924 1924	×××	?	×××	×				×			×		×							
48b	1 ft. 8 in.	Oct. Feb. Apr.	1923 1924 1924	×××		×××	×																
47	2 feet	Oet. Feb. Apr.	1923 1924 1924	××	××						×		××										
46	2 ft. 3 in.	Oct. Feb. Apr.	1923 1924 1924	×××		×××			×		×											×	
45a	2 ft. 6 in.	Oct. Feb. Apr.	1923 1924 1924	×××		×			××														
45b	2 ft. 6 in.	Oct. Feb. Apr.	1923 1924 1924	×××		×						×			×								
44a	3 feet	Oct. Feb. Apr.	1923 1924 1924	×××	××						××		×									×	
44b	3 feet	Oct. Feb. Apr.	1923 1924 1924	×××		×××							N.										
43a	3 ft. 1 in.	Oct. Feb. Apr.	1923 1924 1924	×××	?	×××	×																

# TABLE III (Continued)

Number of culture	Depth of sample	Date of examination	Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Trochiscia sp.	Chlamydomonas (unidentified)	Dactylococcus sp.	Species A	Species B	Species D	Species E	Gloeocystis-like stage	Nostoc commune	Nostoc sp.	Phormidium tenue	Phormidium molle	Navicula atemoides	Nitzschia palea
43b	3 ft. 1 in.	Oct. 1923 Feb. 1924 Apr. 1924	IJХ			×××							×									
43c	3 ft. 1 in.	Oct. 1923 Feb. 1924 Apr. 1924	L)				××															
42a	3 ft. 3 in.	Oct. 1923 Jan. 1924 Apr. 1924	£I .			×××							×××									
42b	3 ft. 3 in.	Oct. 1923 Feb. 1924 Apr. 1924	X X X	? ×		×					×					×					×	
42c	3 ft. 3 in.	Oct. 1923 Feb. 1924 Apr. 1924	Ł									×××										
41a	3 ft. 6 in.	Oct. 1923 Feb. 1924 Apr. 1924	X X X X										××	×								
41b	3 ft. 6 in.	Oct. 1923 Feb. 1924 Apr. 1924	XXX																		×	
41c	3 ft. 6 in.	Oct. 1923 Feb. 1924 Apr. 1924	3 ×										×									
40a	3 ft. 9 in.				Pre	odu	ıce	d n	o į	ro	wtł	1										
<b>4</b> 0b	3 ft. 9 in.	Oct. 1923 Feb. 1924 Apr. 1924	X L L X					×														
40c	3 ft. 9 in.	Oct. 1923 Feb. 1924 Apr. 1924	Ł X			×		×	×													

TABLE IV (Series D) SAMPLES TAKEN IN "LOWER" GARDEN BEHIND GREENHOUSES, JANUARY 14, 1924. (TEMP. 22° F.)

								7 TAT 1			F.,															
Number of culture	Depth of sample	Weight of inoculum	Date of examination	Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Stichococcus bacillaris	S. scopulinus	Ulothrix subtilis	Uronema confervicola	Species B	Species C	Species E	Species F	Species G	Species H	Species I	Species J	Moss Protonema	Nostoc comminutum	Oscillatoria amphibia	Navicula atemoides	Nitzschia palea
77	6 in.	5 gm.	Feb. 1924 Apr. 1924	××	×				××		×	××				×						×		_		
76i	12 in.	.01 gm.	Mar. 1924 Apr. 1924	×					××																	
76h	12 in.	.02 gm.	Mar. 1924 Apr. 1924						×																	_
76g	12 in.	.05 gm.	Mar. 1924 Apr. 1924	×					×					×												_
76a	12 in.	.1 gm.	Feb. 1924 Apr. 1924	×	Γ		×		×													_			×	_
76b	12 in.	.2 gm.	Feb. 1924 Apr. 1924	××	Γ		×		××					×												
76c	12 in.	.5 gm.	Feb. 1924 Apr. 1924	××	Γ				××	_				×												
76d	12 in.	1 gm.	Feb. 1924 Apr. 1924	×	$\lceil$		×		×		_			×				_								_
76e	12 in.	2 gm.	Feb. 1924 Apr. 1924	××		×	×		×		××			×				Į,								
76f	12 in.	5 gm.	Feb. 1924 Apr. 1924	××		×	×			×	×						××	×		_					×	
75	1 ft. 6 in.	5 gm.	Mar. 1924 Apr. 1924	××			××		×			×		×												_
74a	2 feet	.1 gm.	Mar. 1924 Apr. 1924									-						××								_
74b	2 feet	.2 gm.	Mar. 1924 Apr. 1924	×			×				_							_	-			_				_
74c	2 feet	.5 gm.	Mar. 1924 Apr. 1924		×									?												
74d	2 feet	1 gm.	Mar. 1924 Apr. 1924				×		×		-	-						×								
74e	2 feet	2 gm.	Mar. 1924 Apr. 1924	××					×					××		_									×	
74f	2 feet	5 gm.	Mar. 1924 Apr. 1924	×	×	×	×		×					×		×										_

# TABLE IV (Continued)

Number of culture	Depth of sample	Weight of inoculum	Date of examination	Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Stichococcus bacillaris	S. scopulinus	Ulothrix subtilis	Uronema conferricola	Species B	Species C	Species E	Species F	Species G	Species H	Species I	Species J	Moss Protonema	Nostoc comminutum	Oscillatoria amphibia	Navicula atemoides	Nitzschia palea
73	2 ft. 6 in.	5 gm.	Mar. 1924 Apr. 1924	۱× ۱×			×		×	×				×			×								×	
72	2 ft. 9 in.	5 gm.	Mar. 1924 Apr. 1924	¥×		×	X	×	×								8								×	3
71a	3 feet	.1 gm.	Mar. 1924 Apr. 1924											×												
71b	3 feet	.2 gm.	Mar. 1924 Apr. 1924	1											X											
71c	3 feet	.5 gm.	Mar. 1294 Apr. 1924	1 ×							_			×												
71d	3 feet	1 gm.	Mar. 192 Apr. 192	1			×		×			×	×	×					×				-			
71e	3 feet	2 gm.	Mar. 192- Apr. 192-	1 ×	×			_		_			×	×		××			×					·		
71f	3 feet	5 gm.	Mar. 192 Apr. 192		×		×		×			××		×		×				_						
70a	3 ft. 6 in.	5 gm.	Mar. 192 Apr. 192	4																			×			Ī
70b	3 ft. 6 in.	over 5 gm.	Mar. 192 Apr. 192	4 ×	×	×		×						-	?											Ī
69a, b, c	4 ft. 2 in.	.5 gm., 1 gm., and 2 gm. re- spectively				P	roc	luc	ed	no	gr	ow	th													
69d	4 ft. 2 in.	5 gm.	Mar. 192 Apr. 192															X								
69e	4 ft. 2 in.	over 5 gm.	Mar. 192- Apr. 192-											×				×								×
68a, b	5 ft. 5 in.	.5 gm. and 1 gm. resp.	July 1924	ı×																						
68c	5 ft. 5 in.	2 gm.	Mar. 1924 Apr. 1924	1																Ti-					×	
68d	5 ft. 5 in.	5 gm.	Mar. 1924 Apr. 1924																							

# TABLE IV (Continued)

																				_							
Number of culture	Depth of sample	Weight of inoculum	Date of examination		Chlorococcum humicola	Chlorochytrium paradoxum	Protosiphon botryoides	Chlorella sp.	Botrydiopsis arhiza	Trochiscia reticularis	Stichococcus bacillaris	S. scopulinus	Ulothrix subtilis	Uronema conferricolu	Species B	Species C	Species E	Species F	Species G	Species H	Species I	Species J	Moss Protonema	Nostoc comminutum	Oscillatoria amphibia	Navicula atemoides	Nitzschin palea
68e	5 ft. 5 in.	over 5 gm.	Maı. Apr.	1924 1924	×						_				×				_								_
67a	5 ft. 8 in.	.5 gm.	May	7 1924									_								×	5.3					
67b -c	5 ft. 8 in.	1 gm., 2 gm., 5 gm. and over, respec- tively						P	roc	luc	ed	no	gr	оw	th												
66	6 ft. 4 in.	over 5 gm.						P	ro	luc	ed	no	gı	·ow	th												
65a- c	6 ft. 10 in.	.5 gm., 1 gm., and 2 gm. re- spectively						P	roc	luc	ed	no	, KI	ow	rth												
65d	6 ft. 10 in.	5 gm.		1924 1924						-,									×								
65e	6 ft. 10 in.	over 5 gm.				-		P	roc	luc	ed	no	gı	ow	th												
64	7 ft. 6 in.	over 5 gm.						P	roc	luc	ed	no	, ki	row	rth												
63a- d	8 ft. 2 in.	.5 gm., 1 gm., 2 gm. and 5 gm. respec- tively						P	roc	luc	ed	no	gı	row	rth												
63e	8 ft. 2 in.	over 5 gm.	Apr.	1924				3		.,															×		
62	8 ft. 7 in.	over 5 gm.	Apr.	1924				-													×	×					
61a- c	9 ft. 1 in.	1 gm., 2 gm., 5 gm. respec- tively						P	ro	duc	ed	no	g	row	vth												
61d	9 ft. 1 in.	over 5 gm.	Mar.	1924											×								ĺ				
60a- c	9 ft. 5 in.	1 gm., 2 gm., and 5 gm. re- spectively.						F	ro	duc	ed	no	g	row	vth												

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#### PSEUDO-FERTILITY IN NICOTIANA<sup>1</sup>

#### FANNY FERN SMITH

Rufus J. Lackland Research Fellow, Missouri Botanical Garden

#### Introduction

It has long been known that the eggs of certain hermaphroditic plants and animals cannot usually be fertilized by pollen or sperms from the same individual, and this phenomenon has been termed self-sterility. Although it has been demonstrated in only one animal, Ciona intestinalis, the phenomenon has been shown to be fairly widespread throughout the plant kingdom. In 1895, Knuth listed 134 observed self-sterile species of Angiosperms belonging to 46 families, and East and Park ('17) estimated that 70 per cent of these observations proved definitely that the species were self-sterile. Hence, as early as 1895, at least 100 self-sterile species of Angiosperms had been observed.

In Nicotiana, the primary difference between sterile and fertile combinations has been shown (East and Park, '18) to be the difference in rate of pollen-tube growth. In incompatible combinations, the pollen grains germinate but do not grow with sufficient rapidity to reach the ovary before the flower falls. showed, through cytological studies of styles taken at successive twelve-hour intervals after pollination, that the pollen-tubes of fertile combinations exhibit an acceleration in their rate of growth. their growth curves when plotted being similar to those representing autocatalytic reactions. On the other hand, the pollen-tubes of sterile combinations maintain a constant rate of growth, showing no acceleration, their growth curves being straight lines. virtue of the acceleration in the rate of growth, the pollen-tubes of compatible matings reach the ovary in less than 96 hours after pollination, whereas in incompatible matings they fail to reach the ovary within the life of the flower.

<sup>1</sup> An investigation carried out at the Missouri Botanical Garden in the Graduate Laboratory of the Henry Shaw School of Botany of Washington University and submitted as a thesis in partial fulfillment of the requirements for the degree of master of science in the Henry Shaw School of Botany of Washington University.

Cross-incompatibility among self-sterile individuals was not described in the early work on self-sterility, and it was not until the work of de Vries ('06) on the self-sterile species Linaria vulgaris that the existence of intra-sterile, inter-fertile classes was first observed. That is, de Vries demonstrated the fact that there were among the self-sterile individuals of Linaria vulgaris with which he was working, two classes, and that every individual belonging to one class was cross-sterile with every individual of the same class and cross-fertile with every individual of the other Since that time, such intra-sterile, inter-fertile classes have been observed in various species by Correns ('12, '16), East and Park ('17), Baur ('19), Lehmann ('19), Crane ('23), Shull ('23), and Anderson ('24). East and Park ('17) showed that this cross-sterility is identical in nature with self-sterility, and that an interpretation which holds for one phenomenon must likewise hold for the other.

However, among self-sterile plants, individuals exhibiting self-fertility to some degree are frequently found. East and Park ('17) occasionally obtained, in their self-sterile hybrids between Nicotiana alata and N. Forgetiana and in several self-sterile species of Nicotiana, small capsules which contained relatively few seeds and rarely, if ever, approximated the size of those resulting from fertile combinations. It has been agreed by all investigators of the subject that a plant is self-fertile if it sets a full capsule of seeds when pollinated with its own pollen, and self-sterile if no seeds are set after self-pollinations. Neither of these two categories, however, includes those individuals of self-sterile species which exhibit a small but varying amount of self-fertility, and such plants have been described by East and Park as being "pseudofertile."

This phenomenon has been described, although in some cases not by this name, by Correns ('12, '13, '16) in the self-sterile species Cardamine pratensis and Linaria vulgaris; East and Park ('17, '18) in several species of Nicotiana and in hybrids between Nicotiana alata and N. Forgetiana; Sutton ('18) in plums and cherries; Baur ('19) in Antirrhinum hispanicum; Lehmann ('19) in Veronica syriaca; Shull ('23) in Bursa grandiflora; Anderson ('24) in Nicotiana alata, N. Forgetiana, and hybrids between N.

alata and N. Forgetiana; and Smith ('24) in hybrids between N. alata and N. Forgetiana.

East and Park ('17) observed the occurrence of pseudo-fertile individuals particularly among plants which were nearing the end of the flowering season, and hence exhibiting weakened vigor. By comparing the curve of pollen-tube growth in a pseudo-fertile combination with that in a self-sterile mating, East and Park ('18) found indication of the possibility that the so-called phenomenon of pseudo-fertility was a variant of self-sterility which was brought about by unfavorable environmental conditions and general decrease in the vigor of the plant. That is, there was apparently little or no acceleration in the rate of growth of the pollentubes such as is found in normal fertile combinations, but the growth was merely more rapid throughout the course of the pollentube through the style.

Stout ('16, '17), in connection with his studies on self- and crosspollinations in Cichorium Intybus, observed a considerable variation in the expression of self-sterility and attributed it to germinal disturbances, which, however, he concluded were too variable to permit of any type of Mendelian interpretation. This feeble or "partial compatibility," as he terms it, he believes "manifests itself quite indiscriminately throughout the entire period of bloom" and is independent of any decrease in vegetative vigor. Hence, according to him, end-season fertility is comparatively rare and is not a condition commonly operating in incompatible It is quite possible that he was observing in Cichorium Intybus the same phenomenon which was termed "pseudo-fertility" by East and Park ('17) in their work on Nicotiana hybrids, and that he has failed to distinguish between pseudo-fertility and true self-fertility. That is, it is possible that the "sporadic development of self-compatibility giving self-fertility among the progeny of self-sterile lines of descent" which he observed may be due to other changes rather than germinal disturbances. ever, it is true that if the variations which he finds among selfsterile individuals are expressions of pseudo-fertility rather than of true fertility, the degree of pseudo-fertility must be much higher and much more variable in Cichorium Intybus than it is in Nicotiana alata, N. Forgetiana, and hybrids between them.

Stout ('20) reported results of continued work on Cichorium Intybus and of preliminary work on other genera. In Cichorium Intybus, he failed to find any progressive seasonal increase in pseudo-fertility. In Brassica pckinensis he observed a few cases of mid-seasonal fertility. Eschscholtzia californica was found to be somewhat pseudo-fertile, one plant in 200 setting a capsule comparable to those resulting from fertile pollinations, whereas Raphanus sativus was still more self-sterile, only one plant in 200 showing any indication of pseudo-fertility.

Sutton ('18) concludes that in plums and cherries the results are "consistent with the supposition that the plants consist of two larger classes, self-fertiles and self-steriles, with a smaller number of plants of intermediate properties." For this smaller group, she offers two possible interpretations. The intermediate group and some of the self-fertiles may be supposed to be heterozygous, and the self-steriles, homozygous, the occasional indications of partial self-fertility among the latter being attributable to errors probably. On the other hand, when a few fruits are formed out of a large number of pollinated flowers, the fact may mean that compatibility exists in a very slight degree. "If this could be confidently asserted," she adds, "it would be tempting to suppose that the tree may be a mosaic in that respect."

It is this phenomenon of pseudo-fertility, defined by East and Park ('17) and described by numerous investigators before and since, with which the present paper is concerned. In the course of this investigation on Nicotiana alata and on hybrids between N. alata and N. Forgetiana, a comparative measure of pseudofertility has been developed, by means of which it has been possible to demonstrate that pseudo-fertility in self-sterile matings is of the same order as that in cross-sterile matings, that pseudo-fertility is of a different order from true fertility but of the same order as self-sterility, and that in genetic strains concerned, the phenomenon was but slightly affected by environmental changes, or with progress of the flowering season. Measurements of the pollen grains were made and indications were found that the percentage of variability in diameter is significantly greater in plants resulting from self-pollinations than in those coming from cross-pollinations.

#### MATERIALS AND METHODS

In the present series of investigations on the phenomenon of pseudo-fertility, Nicotiana alata Lk. and Otto var. grandiflora Comes, and F<sub>7</sub> and F<sub>8</sub> hybrids between N. alata and N. Forgetiana were used, the seed coming originally from stock used in investigations on self-sterility at the Bussey Institution of Harvard University (see East, '15; East and Park, '17; and Anderson, '24). Strains of the pure species and of the hybrids coming from seeds planted in September, 1922, were taken over by the author in January, 1923, and during the next 18 months the following investigations were conducted on that and the succeeding generation of plants.

The material was particularly satisfactory for such a series of investigations. Capsules containing, on an average, between 300 and 500 seeds each were set in 95 per cent of the compatible combinations; and thus pseudo-fertility, in which usually only a few seeds are set at more or less infrequent intervals, was readily distinguished from true fertility. The plants grow well under greenhouse conditions, and they can be cut back and made to pass through a second flowering season, if so desired. However, due to the attacks of mosaic and to the extremely hot summers, it is almost impossible to carry the plants over from one season to the following in this climate.

Experimental error due to the contamination of pollen or to accidental pollination of flowers was reduced as far as possible. With the anthesis of the first few flowers of a plant, the panicle was enclosed in a paper bag so as to prevent contamination or cross-pollination by wind or insects. When the plants were unbagged for the purpose of pollinating the flowers, care was taken that pollen from one plant was not permitted to fall on the flowers of near-by plants. In making pollinations, newly opened flowers were used in order that there might be as little danger as possible of contamination by foreign pollen. The pollinations were effected by carefully dusting the stigma of a given flower with the desired pollen. When necessary, the flowers were emasculated before being pollinated. Also, after each emasculation or pollination the hands and forceps were washed in 95 per cent alcohol in order to kill all the pollen. That this procedure

was effective in the prevention of contamination by foreign pollen was demonstrated by Anderson ('24), as follows: The fingers were dusted with pollen and 4 pollinations made. Then, after rinsing the hands with alcohol, 4 more pollinations were made on the same plant with the remaining pollen. Full capsules were set in the first 4 pollinations and none in the last, thus showing that the alcohol was efficient in destroying the pollen grains.

# Presentation of Data and Discussion SELF-STERILITY, SELF-FERTILITY, AND PSEUDO-FERTILITY

As has already been stated, the object of these investigations has been a study of pseudo-fertility, particularly with respect to the relation which it bears to self- and cross-sterility, and cross-fertility. It has been indicated by East and Park ('18), through a study of the relative rates of pollen-tube growth, that pseudo-fertility in *Nicotiana* is of the nature of true sterility rather than of that of true fertility. That is, their data indicated that the phenomenon was probably a variant of true sterility, brought about by unfavorable environmental conditions, and not a modified expression of true fertility resulting from germinal modifications in a self-sterile race. The present series of investigations was designed to prove definitely, by a method other than that of pollen-tube growth, that, in *Nicotiana*, pseudo-fertility is of the order of self-sterility and not of self-fertility.

The method used centers about the fact which had previously been observed by Anderson that pollinated unopened buds will frequently exhibit pseudo-fertility when mature flowers will not. With this idea in mind, series of pollinations were made simultaneously on unopened buds, and first and second flowers of the same branch of the panicle (fig. 1). The flowers were numbered from apex to base of the branch of the panicle, thus making the first the youngest of the mature flowers. The series of pollinations were made and the panicles bagged. After one week, those flowers, the ovaries of which showed no enlargement or indication of setting seed, were removed. This was justified by the previously determined fact that certain indication of capsule formation appears within the first week after pollination. Those flowers which indicated that seeds were being set were allowed to

remain on the plant until the capsule showed signs of dehiscence, at which time they were removed. When thoroughly dry, the seeds in each capsule were counted and recorded. As there was not sufficient time to allow those capsules formed during the latter part of the work to mature on the plant, they were removed two

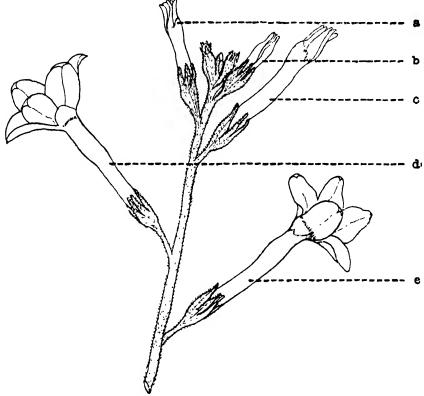


Fig. 1. Typical branch of a panicle: a, young bud; b, very young bud; c, unopened bud; d, first flower; e, second flower.

weeks after the pollinations were made and were dried in an oven at 100° C. for 30 minutes. This heating served to dry the seeds so that they could be readily separated and counted, and was possible by virtue of the fact that the chief aim was a determination of the number of seeds set, and that the seeds were not needed for the plants of the next season.

Such series of pollinations were made on both the hybrids and the pure species, *Nicotiana alata*, but at this point only those results obtained from the hybrids are of interest. Two hundred thirty-three series, including self-sterile, cross-sterile, and cross-fertile combinations were made on the hybrid plants. For the resulting data see "Complete Data" at the close of this paper. In several instances, capsules were formed and were lost before the seeds were counted (represented in the "Complete Data" by "?"), but in all other cases, the seeds of every capsule were counted and recorded. All of the series are included in the "Complete Data," but any series containing one or more such cases has been excluded from the following calculations.

In table 1, the average number of seeds set in the self-sterile, cross-sterile, and cross-fertile combinations respectively are given and these results are graphically presented in fig. 2.

TABLE I
PSEUDO-FERTILITY VERSUS TRUE FERTILITY

Pollinations	No. of series	Average nu	mber of seeds	oer capsule
1 Ollinautolis	110. OI BEITES	Unopened bud	1st flower	2nd flower
Self-sterile Cross-sterile Cross-fertile	84 63 41	86.36 138.65 439.46	27.92 32.16 465.83	5.63 18.88 414.80

Figure 2 expresses several fundamental relations. In the first place, it shows quite clearly that just as cross-sterility was demonstrated by East and Park ('17) to be of the same nature as self-sterility, so here, the expression of pseudo-fertility in cross-sterile combinations is comparable to that in self-sterile combinations. The two curves representing pseudo-fertility in self- and cross-sterile combinations are essentially the same and are in relatively the same position on the graph. The only interpretation which can be applied to this close similarity between these two curves is that pseudo-fertility in cross-sterile combinations is of the same order as that in self-sterile pollinations.

The second and probably the most fundamental relation to be obtained from table I and the curves in fig. 2 is that pseudofertility is not of the nature of true fertility. The curve representing true fertility is not only of a different type from those representing pseudo-fertility, but it occupies an area on the

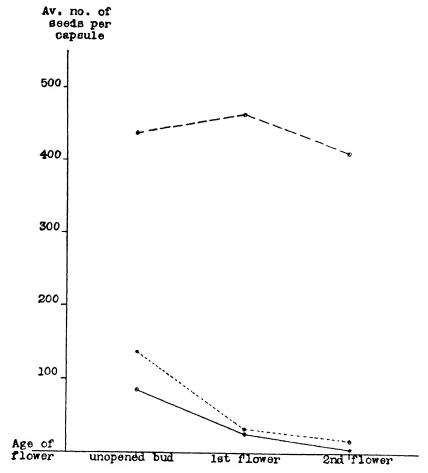


Fig. 2. Pseudo-fertility versus true fertility:——, self-sterile pollinations;————, cross-fertile pollinations.

graph which is widely separated from that occupied by the curves representing pseudo-fertility. In effect, the lowest point on the curve of the former is practically three times as high as the highest point of either of the latter two curves. It is indeed quite obvious that pseudo-fertility as expressed in the hybrids between *Nicotiana alata* and *N. Forgetiana* cannot be of the same order as true fertility.

Just as it is obvious even from a casual examination of fig. 2 that pseudo-fertility in either self- or cross-sterile combinations

is not of the order of complete compatibility, it is likewise quite clearly shown that it is comparable to, and probably a variant of, complete incompatibility. In the graph, the ordinate represents the number of seeds set. Therefore, the curve of truly sterile combinations, or combinations in which no seeds are set, would be a straight line coincident with the abscissa. Hence, to compare pseudo-fertility with complete incompatibility, it is necessary merely to compare the two curves with the abscissa. Such a comparison indicates rather decidedly that the relationship of pseudo-fertility is with complete incompatibility and not with complete compatibility.

A study of the curves themselves is somewhat significant. In the case of truly fertile combinations, one would expect pollinations of first flowers to yield the most abundant seeds, since the second flowers would probably be so old that they would wither before a sufficient number of pollen-tubes reached the ovary to effect fertilization of all the ovules, whereas in the unopened bud probably not all the ovules would be sufficiently well developed to permit of fertilization. Hence, one would expect the curve of fertile pollinations to rise from the unopened bud to a maximum in the first flower and fall again in the second flower. This is exactly the type of curve that has been obtained from an average of 41 cross-fertile combinations.

On the other hand, since self-sterility (according to East, '15) is due to the fact that pollen-tubes after self-pollination show no acceleration in growth and hence fail to reach the ovary within the life of the flower, it would be expected that pollinations of unopened buds would yield the most seeds by virtue of the fact that in such pollinations additional time is gained and the pollentubes may reach the ovary before the flower falls. That this is true, is quite evident in the curve which shows a sudden drop from the unopened bud to the first flower followed by a gradual decline to the second flower. This is further evidence that the pseudofertility of self- and cross-sterile combinations is not of the same order as true fertility.

In this connection, observations made on a single plant, WA-1, are of interest. This plant was a *Nicotiana alata*  $\times$  *N. Forgetiana* hybrid of a genetic strain other than that to which the remainder

of the plants used belonged and hence not included in the "Complete Data." It was apparently completely self-sterile, no seeds being set in 17 series of pollinations made on unopened buds, first and second flowers. Series were pollinated at intervals throughout the flowering season of the plant, which, in this case, extended over a period of 50 days, and there was no evidence whatsoever of an end-seasonal pseudo-fertility. It was, however, possible to obtain seeds in considerable numbers by pollinating buds from 4 to 6 days before anthesis, as shown in table II. Hartley ('02), in attempting premature fertile pollinations in Nicotiana Tabacum, was not only unable to get the flowers to set seed, but also found that the growth of the pollen-tubes into the ovaries before the ovules were sufficiently mature for fertilization resulted in an injury which caused the flowers to fall immediately. In view of his observations, it is interesting that in these sterile combinations, seeds can be set in considerable numbers by pollinating the very young buds.

In the light of this comparison between the curves representing true fertility and the degree of pseudo-fertility as indicated by the number of seeds set in the unopened bud, first and second flowers. we might consider the number of seeds set in younger buds and In cross-fertile combinations seeds have been set older flowers. in the third and fourth flowers in the few cases where they have been tried, but always fewer than in the second flower. fore, as the flower ages, fewer and fewer seeds are set until theoretically we reach an age at which no seeds are set. At this place, the curves representing true fertility and pseudo-fertility would be coincident with each other and with the abscissa. we can conceive of a point at the opposite end of the curves where they would pass through the same point. As younger and younger buds are pollinated in sterile combinations, more and more seeds are set and the curve for pseudo-fertility rises. As will be shown in the case of WA-1, plants which are apparently completely self-sterile when pollinated in the unopened bud, first and second flowers may be pseudo-fertile to a considerable degree if pollinations on younger buds are made. However, in cross-fertile matings, fewer seeds were set in unopened buds than in the first flowers. As younger buds would be pollinated, it seems probable

that fewer and fewer seeds would be set. In effect, Hartley ('02) found in *Nicotiana Tabacum* that very early fertile pollinations caused the flowers to fall before any seeds were set. Hence, we can conceive of the curve for true fertility falling off rather abruptly, and in the drop it would probably cross the rising curve for pseudo-fertility. Therefore, if the two curves would be carried out sufficiently far in either direction it is very probable that they would meet. This point should be considered in any attempt to distinguish between pseudo-fertility and true fertility, and the phenomena should be studied on flowers at that age at which the difference is greatest.

TABLE II
PSEUDO-FERTILITY IN PREMATURE POLLINATIONS

Pollination	Date	No.	of seeds per cap	sule
1 omnation	Date	Very young bud	Young bud	Unopened bud
WA-1 selfed	10-24-23	0	0	0
WA-1 selfed	10-24-23	150+	0	0
WA-1 selfed	10-24-23	37	0	0
WA-1 selfed	10-24-23	0	0	0
WA-1 selfed	10-24-23	432	0	0
WA-1 selfed	11- 3-23	319	0	0
WA-1 selfed	11-3-23	352	0	e
WA-1 selfed	11- 3-23	487	0	0
WA-1 selfed	11-12-23	0	6	0
WA-1 selfed	11-19-23	0	454	0
WA-1 selfed	11-19-23	440	0	0
WA-1 selfed	11-24-23	0	391	0
WA-1 selfed	11-24-23	167	0	0
WA-1 selfed	12- 7-23	0	0	0
WA-1 selfed	12- 7-23	0	150	0
WA-1 selfed	12-13-23	0	0	0

In the table, buds before anthesis are termed young buds and those 6 days before anthesis are termed very young buds. It is of interest to find that seeds can be set by pollinations made so early in the bud, and the fact may prove to be of practical significance in obtaining seeds from normally incompatible combinations. However, before its practical significance could be asserted, it would be necessary not only to run germination tests on the seeds produced by such early pollinations to see whether or not they are fertile, but to discover the extent to which it is possible to obtain seeds by this means from other Angiosperms.

As has been said earlier, the panicles were placed in paper bags so as to avoid cross-pollination by wind or insects. In view of the fact that environmental factors have been thought to increase pseudo-fertility, the question arose as to whether the bagging of the panicles in any way influenced the degree of pseudo-fertility expressed by the plant. In order to determine any such possible influence, the following experiment was carried out.

Two series were made simultaneously on different branches of the same panicle of the plant. The branch bearing one series was bagged, whereas that bearing the other was not included. To avoid accidental cross-pollination in the case of those not bagged, the corollas of the flowers were tied, and in that way no foreign pollen could reach the stigma. Ten such series, including both self- and cross-sterile combinations, were made on different plants, the data of which are given in table III.

TABLE III
INFLUENCE OF BAGGING ON PSEUDO-FERTILITY

		Number of seeds per capsule					
Pollination	Date	Unopened bud		1st flower		2nd flower	
		Bagged	Unbagged	Bagged	Unbagged	Bagged	Unbagged
CA-4×CB-13	4-19-23	338	158	0	0	85	n
CC-1×CC-5	4-26-23		461	390	381	338	261
CC-5×CC-1	4-26-23		0	0	?	0	?
CB-4×CG-8	4-19-23		292	87	105	41	0
CG-20×CL-4	4-19-23	339	0	0	0	0	0
CC-1 selfed	4-26-23	0	0	0	0	0	O
CC-5 selfed	4-26-23	0	0	0	0	0	0
WA-1 selfed	10-19-23	0	0	0	0	0	0
$WA-1 \times WB-1$	10-19-23	0	0	0	0	0	0
WB-1 selfed	10-20-23	0	0	0	0	0	0
Average		140 2	101.22	53	54	52.6	29

<sup>\*</sup> This series not included in the averages because of the lost capsules in the unbagged first and second flowers.

A comparison of the average number of seeds set in the bagged series with that in the unbagged, together with a comparison of their respective graphs (fig. 3), shows quite clearly that bagging does increase the degree of pseudo-fertility. Considering together the average number of seeds set in unopened bud, first and second flowers, there has been an increase of 33.42 per cent in the

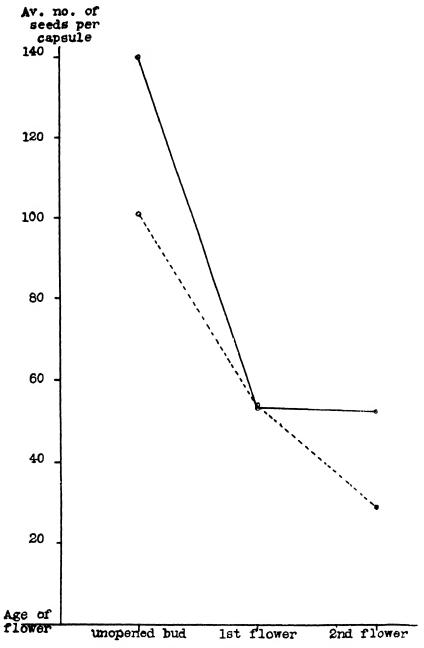


Fig. 3. Influence of bagging on pseudo-fertility: ———, bagged flowers; unbagged flowers.

number of seeds set in the bagged over those in the unbagged panicles. The fact that pseudo-fertility is thus increased by bagging must be remembered in considering the actual values of the data as given. However, bagging was practised throughout the entire series of pollinations and hence the relative values still remain true. The error resulting from bagging the flowers is constant throughout the experiments and hence does not affect the conclusions which are to be drawn from the data recorded.

#### INFLUENCE OF SEASONAL PROGRESS

East and Park ('17) suggested that pseudo-fertility was a variant of self-sterility brought about by unfavorable environmental conditions which caused a decrease in the vigor of the plant. Particularly did they find pseudo-fertility near the end of the flowering season. With this idea in mind, successive series of self-pollinations were made on 16 individuals during the early, and then during the latter part of their flowering season. The flowering season of the plants grown in the Bussey Institution was sometimes as long as 3 months, but in our greenhouses this was not the case. Only rarely did the flowering season of any one plant continue for more than 3 weeks, and usually it lasted only 10 days or 2 weeks. Hence, with the exception of the plant WA-1 previously cited, the interim between the early and later pollinations was a week to 18 days. The data from these repeated series are given in table IV.

TABLE IV
SEASONAL CHANGE IN PSEUDO-FERTILITY

Pollination	Date	Number of seeds per capsule			
1 omnation	Dave	Unopened bud	1st flower	2nd flower	
GOB-4 selfed GOB-4 selfed	4-21-24 4-30-24	0 314	0	0	
GA-2 selfed GA-2 selfed	4-18-24 4-30-24	0 27	0	0	
GC-3 selfed GC-3 selfed	4 -14 -24 4-26-24	124 387	0	0 <b>0</b>	
GC-6 selfed GC-6 selfed	4-18-24 4-26-24	0 149	0	0	

TABLE IV (Continued)

Pollination	Date	Number of seeds per capsule		
1 omnation Date		Unopened bud	1st flower	2nd flower
GF-1 selfed	4-16-24	0	o	0
GF-1 selfed GF-1 selfed	4-28-24 4-30-24	329	0	0
	}		_	0
LH-1 selfed LH-1 selfed	4-21-24 4-28-24	67 45	0 79	0
LH-1 selled	4-28-24	45	79	U
CB-14 selfed	3-27-23	473	136	48
CB-14 selfed	4-14-23	550	29	54
GC-1 selfed	3-29-24	48	0	o
GC-1 selfed	4- 8-24	14	0	0
GF-2 selfed	4-16-24	121	0	0
GF-2 selfed	4-28-24	0	Ō	0
GF-9 selfed	4-21-24	0	208	o
GF-9 selfed	4-28-24	ŏ	0	Ŏ
GOA-2 selfed	4-16-24	419	154	0
GOA-2 selfed	4-30-24	7	0	ŏ
GOF-3 selfed	4-18-24	0	0	0
GOF-3 selfed	4-30-24	Ŏ	ŏ	ŏ
GE-1 selfed	4-21-24	0	0	0
GE-1 selfed	4-28-24	0	0	ő
COA 116 3	4-11-24	0	0	0
GOA-1 selfed GOA-1 selfed	4-11-24	0	0	0
GOA-3 selfed	4-18-24	0	0	0
GOA-3 selfed	4-30-24	0	0	0
GOA-4 selfed	4-21-24	0	0	0
GOA-4 selfed	4-30-24	0	0	0

The possible change in degree of pseudo-fertility with seasonal progress is best studied by comparing averages (see table v) of the number of seeds set in the early part of the season with those in the later pollinations. From this table and fig. 4 it appears evident

TABLE V SEASONAL CHANGE IN PSEUDO-FERTILITY

	Average number of seeds per capsule			
	Unopened bud	1st flower	2nd flower	
Early pollinations Later pollinations	78.25 107.625	31.125 6.75	3 3.375	

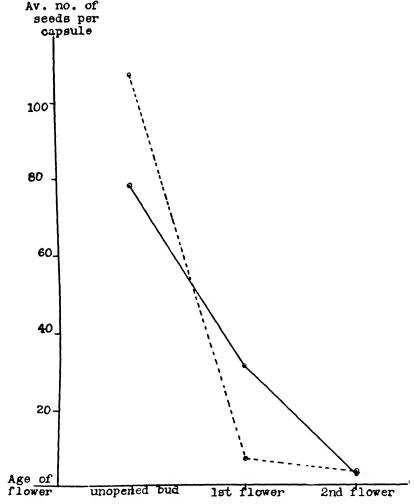


Fig. 4. Seasonal change in pseudo-fertility: ———, early pollinations; ———, later pollinations.

that, considering all 3 pollinations of each series, there was very little change of pseudo-fertility with the progress of the season. The average number of seeds set in the later series is only 4.78 per cent greater than that in the early pollinations and this is scarcely great enough to be significant. Hence, there was little if any seasonal change in pseudo-fertility shown in the *Nicotiana alata*  $\times$  *N. Forgetiana* hybrids used in the present investigations.

#### PSEUDO-FERTILITY IN NICOTIANA ALATA

All of the above investigations were conducted on hybrids which were the seventh and eighth generation descendants of a cross between Nicotiana alata and N. Forgetiana. A few series, including self-sterile, cross-sterile, and cross-fertile pollinations, have likewise been made on several genetic families of the pure species, Nicotiana alata (see "Complete Data" at close of this paper). Although the data obtained here may not be sufficient to permit of positive conclusions, it does seem significant that in 22 series of self- and cross-sterile pollinations not a single seed was set, whereas in the 6 fertile crosses the number of seeds per capsule averaged as follows:

Unopened bud	511.83
First flower	409.5
Second flower	521.67

These results indicate an absence of pseudo-fertility in *Nicotiana* alata, or at least in those strains of that species with which these investigations were carried out.

#### ABNORMAL POLLEN AND SELF-POLLINATIONS

During the course of the investigation, a microscopic examination of the pollen grains was made, and the fact was observed that among the hybrids some plants showed a greater per cent of abnormal pollen than others. The preliminary observations suggested that there was probably some correlation between the per cent of abnormal pollen, the degree of pseudo-fertility, and the amount of self-pollination in the history of the genetic strain concerned. Hence, the pollen grains of the various strains were studied, and in the case of each plant examined the diameters of 50 pollen grains, taken directly across the field of the microscope so as to insure a random sample, were measured by means of an eye-piece micrometer (see "Complete Data" for the resulting measurements).

Pollen of plants containing a considerable amount of small pollen grains likewise contains an approximately equal number of unusually large grains, since, due to the abortion of some of the grains, others are given the opportunity of an unusual degree of development. Hence, in considering the measurements of 50

grains, those plants with a low percentage of abnormal pollen have many grains near the mean diameter, with very few, if any, at the extreme. On the other hand, those plants with a high percentage of abnormal pollen have fewer grains of the mean diameter and more at the extremes than do those with a low percentage. Consequently, the relative per cent of abnormal pollen can be judged by the relative height and spread of the curves, which can in turn be measured by the standard deviation,  $\sigma$ . The standard deviation, then, can be used as a basis for comparison of the amount of abnormal pollen in any two groups of plants, and by this means it is possible to determine whether or not there is a significant difference between the two strains.

Class	Genetic families included		
Selfed of selfed Selfed of crossed Crossed of selfed Crossed of selfed and crossed Crossed of crossed	LA, LC, LE CA, CB, CC, CD GOB GA, GC, GOF, GOX CL, CM, CG, CH, GE, GF, GOA, GOC, LH		

Each group of families was considered as a unit. Frequency tables were made for each of them, and the standard deviation was calculated in every case with the following results.

Class	No. of grains	Standard deviation
Selfed of selfed Selfed of crossed Crossed of selfed	450 475 350	$.9933 \pm .0223$ $1.0834 \pm .0237$ $.7032 \pm .0179$
Crossed of selfed and crossed Crossed of crossed	1500 1300	$.6098 \pm .0075$ $.7296 \pm .0097$

The notation must be made here that the measurements in the selfed of crossed families and in a few of the crossed of crossed families were made early in the work and only 25 grains were measured in each plant. The latter have not been brought into this calculation, but since there were no measurements made on the selfed of crossed families during the latter part of the work, they are included here.

Since it was conceived that possibly the abnormal pollen was correlated with the degree of self-pollination in the history of the plants or with the recency of the self-pollination, the plants were divided into the following groups on this basis. The "selfed of

selfed" class came from seeds which were the result of self-pollinations made on plants which, in turn, were the results of self-pollinations. The "selfed of crossed" class came from self-pollinations on plants which were the product of a cross-pollination. The "crossed of selfed" class were plants which were the product of crosses between plants which, in turn, had come from self-pollinations. The "crossed of selfed and crossed" class were those individuals coming from crosses between two plants, one of which was the result of a self-pollination and the other the product of a cross between two parent plants. Finally, the "crossed of crossed" plants were the product of crosses between plants which, in turn, were the product of cross-pollinations.

To determine the significance of the difference it is simply necessary to divide the difference in the values of  $\sigma$  by the probable error of that difference, and if the quotient is greater than three the difference is significant.

P.E.<sub>diff.</sub> =  $\sqrt{E_{\sigma_1}^2 + E_{\sigma_2}^2}$  where  $E_{\sigma_1}$  = probable error of  $\sigma$  in one group and  $E_{\sigma_2}$  is that of  $\sigma$  in the other. Therefore,

$$\frac{\text{Diff.}}{\text{P.E.}} - \frac{\text{Diff.}}{\sqrt{\mathbf{E_{\sigma_1}}^2 + \mathbf{E_{\sigma_2}}^2}}$$

By means of this equation, the selfed of selfed group was compared with each of the other classes of plants with the following results:

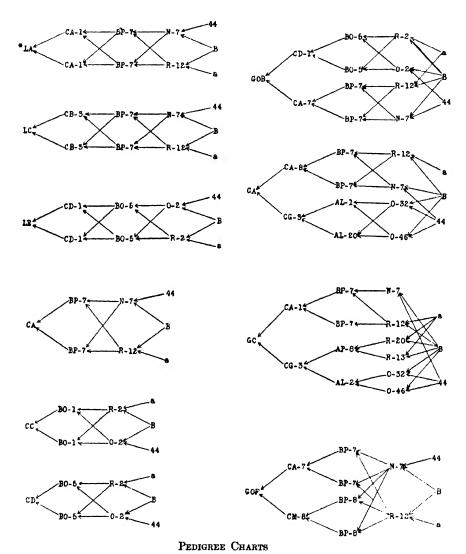
Class	Diff. P.E.
Selfed of crossed	2.77
Crossed of selfed	10 14
Crossed of selfed and crossed	16.32
Crossed of crossed	10.85

These results then indicate that there is no significant difference between the standard deviation in diameter of pollen grains in the selfed of selfed, and selfed of crossed classes. On the other hand, there is a very significant difference between that of the above 2 classes and that of the crossed of selfed, crossed of selfed and crossed, and crossed of crossed groups. As has been shown above, a significant difference in standard deviation indicates a

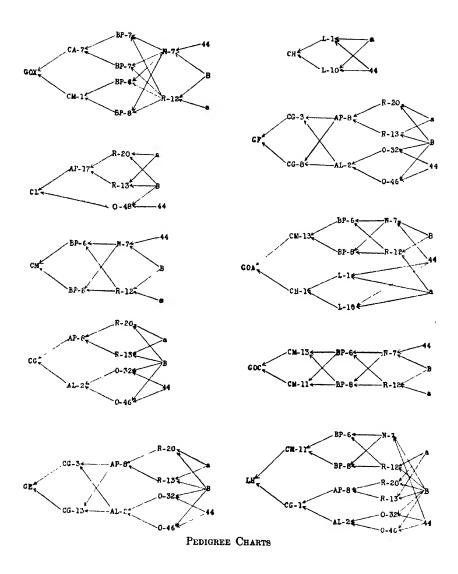
significant difference in the percentage of abnormal pollen. Hence, in the strains studied, it appears that the percentage of poor pollen in plants which are the immediate products of self-pollinations is significantly greater than that in those plants which are the immediate products of cross-pollinations. It is interesting that, although poor pollen is generally considered the result of hybridization, here, among hybrids, self-pollination greatly increases the percentage of abnormal pollen over that found in plants resulting from cross-pollinations.

#### SUMMARY

- 1. In the Nicotiana alata  $\times$  N. Forgetiana hybrids, pseudo-fertility is exhibited in both self- and cross-sterile combinations, and is of the same order in both cases.
- 2. It has been definitely demonstrated that in *Nicotiana* pseudo-fertility is not of the same order as true fertility, but that it does stand in direct relation to true sterility.
- 3. Although in some cases, perhaps, pollinations were not made at the extreme end of the season, no seasonal change in the degree of expression of pseudo-fertility was observed.
- 4. Bagging the panicle was found to increase the degree of pseudo-fertility, but it has been shown that this does not influence the conclusions, since the effect was constant throughout the investigations discussed here.
- 5. In one of the hybrid plants, seeds were obtained by pollinating buds 4 to 6 days before anthesis, when none were set in 17 series of pollinations on unopened buds, first, and second flowers.
- 6. Pseudo-fertility was not exhibited in any of the self- and cross-sterile pollinations on plants of *Nicotiana alata*.
- 7. The results of pollen measurements indicate that there is a larger percentage of abnormal pollen in families arising from self-pollinations than in cross-bred strains.



\* The family LA is the result of a self-pollination on plant 1 of family CA, which, in turn, came from a self-pollination on plant 7 of family BP. The family BP, in turn, is the product of a cross between plant 7 of family N, and plant 12 of family R. These families have come from plants 44 and B, and B and a, respectively.



# TABLE VI COMPLETE DATA NICOTIANA ALATA × NICOTIANA FORGETIANA

LA-1 selfed 4-28-24 0 0 0 LA-3 selfed 4-28-24 0 0 0 LC-1 selfed 4-14-24 37 0	0 0 0 0
LA-3 selfed 4-28-24 0 0 0 LC-1 selfed 4-14-24 37 0	0 0 0
LC-1 selfed 4-14-24 37 0	0 0
LC-1 selfed   4-14-24   37   0	0
	0
LC-2 selfcd 4-14-24 42 0	
LC-9 selfed 4-28-24 40 0	0
CA-1 selfed 3-17-23 518 89 CA-3 selfed 3-30-23 ? 30	0
CB-3 selfed 3-30-23 217 30 7	56
CB-7 selfed 3-31-23 ? 282	26
CB-10 selfed 3-31-23 413 311	27
CB-13 selfed 3-31-23 ? 28	<b>8</b> i
CB-14 selfed 3-27-23 473+ 136+	48+
CB-14 selfed 4-14-23 550 29	54
CC-1 selfed 4-26-23 0 0	0
CC-1 selfed 4-26-23 0 0	0
CC-5 selfed 4-26-23 0 0	0
CC-5 selfed 4-26-23 0 0	Ō
CC-9 selfed 4-3-23 0 0	0
CD-1 selfed 4-5-23 661 0	0 ?
CD-4 selfed 4-5-23 588 27 GOB-1 selfed 4-16-24 387 79	120
GOB-1 selled 4-16-24 387 79 GOB-2 selfed 4-18-24 326 217	0
GOB-2 selled 4-18-24 320 217 GOB-4 selfed 4-21-24 0	ő
GOB-4 selfed 4-30-24 314 0	Ŏ
GOB-6 selfed 4-28-24 491 262	211
GOB-7 selfed 4-21-24 0 168	0
GA-1 selfed 4-21-24 0 0	0
GA-2 selfed 4-18-24 0 0	0
GA-2 selfed 4-28-24 0 0	0
GA-2 selfed 4-30-24 27 0	0
GA-3 selfed 4-28-24 0 0	Ō
GA-4 selfed 4-28-24 284 116	Ö
GA-5 selfed 4-30-24 0 0	0
GA-6 selfed 4-30-24 0 0 GC-1 selfed 3-29-24 48 0	0 0
GC-1 selfed 3-29-24 48 0 GC-1 selfed 4-8-24 14 0	0
GC-1 selled 4-3-24 14 0 0 GC-2 selfed 4-26-24 123 0	Ŏ
GC-3 selfed 4-14-24 124 0	ŏ
GC-3 selfed 4-26-24 387 0	ŏ
GC-4 selfed 4-14-24 62 0	Ŏ
GC-4 selfed   4-16-24   140   0	0
GC-5 selfed 4-14-24 29 0	0
GC-6 selfed 4-18-24 0 0	Õ
GC-6 selfed 4-26-24 149 0	Ŏ
GC-9 selfed 4-26-24 100 0	Ō
GC-12 selfed 4-18-24 0 0	0
GOF-2 selfed 4-21-24 75 0	0
GOF-3 selfed 4-18-24 0 0	0
GOF-3 selfed 4-30-24 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	0 0
GOF-4 selfed 4-28-24 0 103 GOF-4 selfed 4-30-24 4 4	13
GOF-5 selfed 4-21-24 0 0	0

Pollination	Date	Unopened bud	1st flower	2nd flower
GOX-1 selfed	4- 8-24	0	0	0
GOX-1 selfed	4-11-24	0	0	0
GOX-3 selfed	4-30-24	0	0	0
GOX-5 selfed	4-30-24	0	0	0
GE-1 selfed	4-21-24	0	0	0
GE-1 selfed	4-28-24	0	0	0
GF-1 selfed	4-16-24	0	0	0
GF-1 selfed	4-28-24	329	0	0
GF-1 selfed	4-30-24	0	0	0
GF-2 selfed	4-16-24	121	0	0
GF-2 selfed	4-28-24	0	0	0
GF-3 selfed	4-30-24	0	0	0
GF-5 selfed	4-30-24	0	0	O
GF-6 selfed	4-21-24	146	0	0
GF-7 selfed	4-21-24	31	371	0
GF-9 selfed	4-21-24	0	208	0
GF-9 selfed	4-28-24	0	0	0
GF-8 selfed	4-30-24	117	0	0
GOC-1 selfed	5- 1-24	0	0	0
GOC-3 selfed	4-30-24	76	0	0
CH-2 selfed	3-27-23	0	0	0
CM-12 selfed	4-14-23	0	0	0
GOA-1 selfed	4-11-24	0	0	0 0 0 0 0
GOA-1 selfed	4-14-24	0	0	0
GOA-1 selfed	4-30-24	0	0	0
GOA-2 selfed	4-16-24	419	154	0
GOA-2 selfed	4-30-24	7	0	0
GOA-3 selfed	4-18-24	0	0	0
GOA-3 selfed	4-30-24	0	0	Ō
GOA-4 selfed	4-21-24	0	0	0 0 0
GOA-4 selfed	4-30-24	0	0	0
LH-1 selfed	4-21-24	67	0	0
LH-1 selfed	4-28-24	45	79	Ŏ
CG-1 selfed	4- 6-23	0	0	0
CG-5 selfed	3-27-23	24	0	0
CG-9 selfed	4- 7-23	54	0	0
CG-10 selfed	3-27-23	0	0	0
CG-18 selfed	4- 5-23	?	?	0
CG-20 selfed	4- 7-23	?	0	Ō

Cross-sterile pollinations										
Pollination	Date	Unopened bud	1st flower	2nd flower						
CA-3 × CA-1 CA-3 × CA-1 CA-3 × CA-4 CA-13 × CA-11 CA-4 × CB-13 CA-4 × CB-13 CA-6 × CB-4 CA-6 × CB-9 CA-11 × CB-15	4-29-24 3-30-23 3-30-23 3-25-23 4-19-23 4-10-23 4-10-23 3-27-23	72 ? ? 378 338 158 ? 385 379	0 46 69 307 0 0 195 0 231	0 0 0 155 55 0 314 0						
CA-12 × CB-9 CB-7 × CB-5 CB-7 × CB-14 CB-12 × CB-6 CB-13 × CB-10	4-10-23 3-31-23 3-31-23 3-27-23 4- 5-23	250 ? ? ? ?	0 0 98 ?	0 0 ? 0						

Pollination	Date	Unopened bud	1st flower	2nd flower
$CB-13 \times CB-14$	3-31-23	21	0	0
CB-14 × CB-15 CB-14 × CA-4 CC-1 × CC-5	4- 5-23	538	276	0
$CB-14 \times CA-4$	4-14-23	418	31	?
$CC-1 \times CC-5$	4-26-23	355	390	338
$CC-1 \times CC-5$	4-26-23	461	381	261
$CC-5 \times CC-1$	4-26-23	0	l ő	0
$\begin{array}{ccc} \text{CC5} & \times & \text{CC1} \\ \text{CC6} & \times & \text{CC9} \end{array}$	4-26-23	0	?	?
CC-6 × CC-9 CC-7 × CC-9 CC-8 × CC-7 CC-9 × CC-7	4-26-23 4- 3-23	0	164	0
CC-8 × CC-7	4- 3-23	261	0 ?	0
CC-9 × CC-7	4- 7-23	0	Ó	205
$\overrightarrow{CD}$ -1 $\widehat{\times}$ $\overrightarrow{CD}$ -2	4- 5-23	536	ŏ	203
$GA_{-3} \vee GA_{-4}$	4-18-24	000	ŏ	Ò
GA-3 × GA-6 GA-5 × GA-2 GA-6 × GA-4 GA-15 × GA-16 GC-3 × GC-4	4-28-24	Ö	ŏ	Ĭŏ
$GA-5 \times GA-2$	4-28-24	194	ŏ	ŏ
$GA-6 \times GA-4$	4-28-24	257	Ŏ	Ŏ
$GA-15 \times GA-16$		0	Ŏ	Ŏ
$\begin{array}{ccc} GC-3 & \times & GC-4 \\ GC-5 & \times & GC-7 \end{array}$	4-18-24	183	?	0
$GC-5 \times GC-7$	4-21-24	248	0	0
$GC-9 \times GC-4$	4-26-24	0	0	0
$GF-7 \times GF-9$	4-28-24	530	136	47
GC-5 × GC-7 GC-9 × GC-4 GF-7 × GF-9 GF-9 × GF-18	4-30-24	8	0	0
GOC-1 × GOC-3 GOC-3 × GOC-1 CM-8 × CM-3 CM-8 × CM-6 CM-11 × CM-5 CG-2 × CG-10 CG-5 × CG-10 CG-6 × CG-5 CG-6 × CG-8 CG-9 × CG-1 CG-10 × CG-2 CG-10 × CG-2 CG-10 × CG-2 CG-10 × CG-2 CG-10 × CG-2 CG-10 × CG-2 CG-10 × CG-2	4-30-24	0	0	0
$GOC_{-3} \times GOC_{-1}$	4-30-24	63	0	0
$CM-8 \times CM-3$	4- 7-23	461	0 ? 0	0
$CM-8 \times CM-6$	4- 7-23	141	?	0
$CM-11 \times CM-3$	4-14-23	250	0	0
$CM-12 \times CM-5$ $CG-2 \times CG-10$	4-14-23 4- 3-23	0 424	Ü	0
$\begin{array}{ccc} \text{CG-2} & \times & \text{CG-10} \\ \text{CG-5} & \times & \text{CG-10} \end{array}$	3-27-23	7	0 ?	0 ?
$CG-6$ $\times$ $CG-10$	4- 7-23	?	ó	ó
CG-6 × CG-8	4- 7-23	ó	ŏ	23
CG-8 × CG-10	4- 5-23	470	41	32
$\overrightarrow{CG}$ -9 $\times \overrightarrow{CG}$ -1	4- 3-23	0	0	0
$\widetilde{\text{CG}}$ -10 $\widehat{\times}$ $\widetilde{\text{CG}}$ -2	3-27-23	ŏ	ŏ	ŏ
CG-10 × CG-2	4-3-23	ŏ	ŏ	Ö
CG-10 × CG-2 CG-18 × CG-8	4- 7-23	?	Ō	0
$CG-19 \times CG-4$	4 -5-23	0	Ŏ	0
$CL-3 \times CL-5$	4- 7-23	36	0	0
$CL-4 \times CL-10$	4- 5-23	72	0	0
$\begin{array}{ccc} \text{CL-4} & \times & \text{CL-10} \\ \text{GOA-1} & \times & \text{GOA-2} \\ \text{GOA-2} & \times & \text{GOA-4} \\ \text{GOA-3} & \times & \text{GOA-1} \end{array}$	4-23-24	0	ŏ	0
$GOA-2 \times GOA-4$	4-26-24	322	Ō	10
$GOA-3 \times GOA-1$	4-26-24	0	0	0
$GA-5 \times GC-6$	5- 1-24	0	0	0
$GE-1 \times GOF-1$		4	0	0
$\begin{array}{ccc} GF-7 & \times & GC-9 \\ GOA-1 & \times & GE-1 \end{array}$	4-30-24	0	0	0
$GOA-1 \times GE-1$ $GOA-4 \times GOB-1$	4-23-24 4-26-24	11 289	0	0 ?
		120	ő	Ó
$GOA-1 \times GOX-1$ $GOB-1 \times GOA-4$		161	0	Ö
$GOC-1 \times GOA-2$	4-26-24	0	ő	ŏ
$\begin{array}{ccc} GOC-1 & \times & GOA-2 \\ GOC-1 & \times & GOX-1 \\ GOF-5 & \times & GOX-3 \end{array}$	4-26-24	ŏ	ŏ	Ŏ
$GOF-5 \times GOX-3$	4-28-24	ŏ	0	ŏ
(i()X-1 X (i()A-4	4-26-24	ŏ	ŏ	ŏ
$LA-1 \times LC-6$	4-28-24	243	0	ŏ
LH-i $\hat{\times}$ Gi	4-28-24	76	37	409
LH-1 × LA-1	4-28-24	0	o l	0
$LH-1 \times LC-5$	4-28-24	75	0	0
$CB-4 \times CG-8$	4-19-23	230	87	41

Pollination	Pollination Date		1st flower	2nd flower
$CB-4 \times CG-8$	4-19-23	292	105	0
$CC-1 \times CL-4$	4-26-23	? !	413	26
$CC-5 \times CL-4$	4-26-23	0	0	0
$CC-5 \times CL-4$	4-26-23	124	?	0
$CD-4 \times CG-20$	4- 5-23	519	0	0
$CG-20 \times CL-4$	4-19-23	334	0	0
$CG-20 \times CL-4$	4-19-23	0	0	0
$CH-1 \times CM-6$	3-31-23	?	0	0
$CM-12 \times CH-2$	4-14-23	0	0	0
$CG-5 \times CB-10$	3-27-23	0	0	0

# Cross-fertile pollinations

Pollination	Date	Unopened bud	1st flower	2nd flower
LC-4 × LC-3	4-28-24	48	?	216
$\overrightarrow{LC}$ -5 $\times$ $\overrightarrow{LC}$ -1	4-28-24	422	153	187
$\overrightarrow{GOB}$ -7 $\times$ $\overrightarrow{GOB}$ -3	4-28-24	540	?	390
$\widetilde{GA}$ -3 $\times$ $\widetilde{GA}$ -15	4-28-24	371	274	?
GA-6 × GA-8	4-28-24	342	378	285
$\overrightarrow{GA-6} \times \overrightarrow{GA-15}$	4-28-24	357	343	225
$\overrightarrow{GC}$ -5 $\times \overrightarrow{GC}$ -9	4-16-24	385	437	430
$\overrightarrow{GC}$ - $\overrightarrow{9}$ $\times$ $\overrightarrow{GC}$ - $\overrightarrow{3}$	4-26-24	522	$2\overline{2}2$	627
GOF-3 × GOF-1	4-21-24	393	320	279
$\overrightarrow{GOF}$ -5 $\times$ $\overrightarrow{GOF}$ -13	4-30-24	181	245	430
$\overrightarrow{GOX}$ -1 $\times$ $\overrightarrow{GOX}$ -4	4-26-24	359	411	502
$GOX^{-1} \times GOX^{-4}$	4-26-24	288	475	511
$GF-1 \times GF-10$	4-28-24	291	396	379
$\overrightarrow{GF-7} \times \overrightarrow{GF-10}$	4-30-24	528	646	591
$GF-9 \times GF-10$	4-28-24	494	390	369
$CM-1 \times CM-3$	4- 7-23	689	669	000
$CM-1 \times CM-12$	4- 7-23	000	666	ŏ
$CM-1 \times CM-14$	4- 7-23	254	455	321
$CM-11 \times CM-14$	4- 7-23	500	700	409
$CG-3 \times CG-8$	4- 7-23	301	248	243
$CG-5 \times CG-7$	4- 7-23	362	334	410
	4- 3-23	838	969	825
$CG-6 \times CG-1$	4- 5-23	200	530	488
$CG-18 \times CG-9$	4- 5-23	481	515	455
CL-2 × CL-1	4- 7-23	467	458	418
$CL-5 \times CL-10$				713
$CL-8 \times CL-1$	4- 7-23	311	780	724
CL-8 × CL-6	4- 7-23 4- 7-23	787 321	834 396	446
$CL-10 \times CL-2$				703
$GC-3 \times GOA-1$	4-14-24	433	685	295
$GOB-1 \times GA-1$	4-30-24	293	422	101
$GOE-1 \times GOF-1$	4-18-24	381	171	
$GOF-4 \times GOA-4$	4-28-24	234	80	341 327
$CA-3 \times CH-1$	3-30-23	?	673	
$CB-14 \times CH-2$	4-14-23	444	396	484
$CB-14 \times CM-12$	4-14-23	246	256	247
$CB-15 \times CM-12$	3-27-23	315	211	0
$CD-1 \times CA-1$	4- 5-23	1004	873	908
$CD-4 \times CL-5$	4- 5-23	681	398	599
$CD-4 \times CM-6$	4-14-23	257	570	496
$CF-3 \times CH-1$	3-27-23	619	343	515
$CG 20 \times CA-1$	4-7-23	511	577	446
$CH-1 \times CG-18$	3-31-23	757	586	649
$CH-2 \times CA-4$	4-14-23	388	395	?
$CH-2 \times CB-14$	4-14-23	356	377	342
$CH-2 \times CL-6$	4-14-23	440	439	429
$CM-11 \times CG-1$	4-3-23	?	553	?
$CM-12 \times CA-4$	4-14-23	424	646	24
$CM-12 \times CB-14$	4-14-23	750	612	431
$CM-12 \times CL-6$	4-14-23	500	717	627

NICOTIANA ALATA									
Self-sterile pollinations									
Pollination	Date	Unopened bud	1st flower	2nd flower					
CJ-3 selfed	4- 3-23	0	0	0					
HA-3 selfed	4-18-24	0	0	0					
HA-3 selfed	5 1-24	0	0	0					
HA-4 selfed	4-29-24	0	0	0					
HA-5 selfed	4-21-24	0	0	0					
HB-1 selfed	4-19-24	0	0	0					
HC-2 selfed	4-21-24	0	0	0					
HC-2 selfed	4-29-24	0	0	Ö					
HC-3 selfed	4-29-24	0	0	0					
HI-1 selfed	4-8-24	0	0	0					
HI-2 selfed	4-14-24	0	0	0					
HI-3 selfed	4-14-24	0	0	0					
HI-3 selfed	5-1-24	0	0	0					
HI-4 selfed	4-16-24	0	0	0					
HI-5 selfed	4-18-24	0	0	0					
HI-8 selfed	4-21-24	0	0	0					
	Cross	sterile pollination	8						
$CJ-1 \times CJ-3$	4-3-23	0	0	0					
$CJ-2 \times CJ-1$	4- 3-23	0	0	0					
$HA-5 \times HA-6$	4-29-23	0	0	0					
$HC-2 \times HC-1$	4-14-24	0	0	0					
$HC-2 \times HC-1$	4-29-24	0	0	0					
$HI-2 \times HI-9$	4-29-24	0	0	0					
	Cross	fertile pollinations	3						
$HA-1 \times HA-5$	4-29-24	679	600	605					
$HA-5 \times HA-3$	4-29-24	600	663	500					
$HA-6 \times HA-3$	4-29-24	637	737	831					
$HC-1 \times HC-5$	4-29-24	155	446	258					
$HC-2 \times HC-4$	4-29-24	1000	13	936					
$HA-I \times HI-9$	4-18-24	787	654	773					
$HA-5 \times HI-2$	4-24-24	900	1569	1079					
$HB-1 \times HC-1$	4-21-24	450	500	1000					

TABLE VII POLLEN MEASUREMENTS (IN TERMS OF DIVISIONS ON MICROMETER SCALE; 1 DIVISION = 8 MICRA)

Donas, i Bivision B Milani,													
	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0
CA-7 CA-13					4 4		17 16		3 4		1 1		
CB-1 CB-4 CB-5 CB-7	1		2 3 6		4 3 7 6		13 14 8 5		5 3 10 6		1 2 1		
CC-1 CC-2 CC-3 CC-4 CC-5	1		4 1		3 3 1 2 1		11 10 10 14 16		7 4 8 9 5		4 2 3 2		1 2

	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0
CC-8 CC-10 CC-11			2 1 1		3 3 3		7 8 17		7 10 3		6 3 1		
CD-1 CD-2 CD-3 CD-4 CD-8	1 2 1 3 1		5 4 4 6 1		2 3 7 4 4		8 13 13 10 15		9 3 2 4				
CL-4 CL-7 CL-8 CL-11			3		3 9 6 3		8 16 16 22		13		1		
CM-1 CM-6 CM-10 CM-13					2 2 1 2		20 21 19 19		3 2 5 4				
CG-20					9		16						
CH-1					1		22		1			1	
GA-1 GA-2 GA-3 GA-4 GA-5 GA-6 GA-8 GA-15 GA-16			1 1 1 2 2	6 3 4 1 4 3 1 2 3	14 15 8 13 2 7 4 7	20 25 22 23 27 22 20 23 19	8 6 15 10 14 14 24 16 13	1 1 2 2 2 1 2 3					
GC-2 GC-3 GC-4 GC-6 GC-7 GC-9 GC-13	2 3		3 5 5 2	1 7 3 7 1	10 10 15 6 14 10	16 17 11 16 17 10	24 19 14 25 5 19 26	3 2 1					
GE-1	3		2	2	6	17	20						
GF-1 GF-2 GF-3 GF-4 GF-5 GF-6 GF-7 GF-8 GF-9 GF-10 GF-11 GF-18	2 2 1	1	3 3 1 1 1	1 2 3 6 2 2 1 2 1	10 3 5 4 8 4 3 6 6 4 4 3	23 23 13 22 25 15 20 18 28 23 26 24	16 16 26 7 7 25 22 25 16 20 17 20	1 4 4 1 2 2 1	5				
GOA-1 GOA-2		1	1 6	8	8 11	25 12	12 11	1 2					

	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0
GOA-3 GOA-4 GOA-5 GOA-6 GOA-7	8	1 20	3 2 8	2 10 2	10 13 2 1 3	17 15 10 5 8	20 7 21 7 18	1 1 10 19	1 1 1	1	1		
GOB-1 GOB-2 GOB-3 GOB-4 GOB-6 GOB-7 GOB-8	1	2 4 2 2	1 7 7 3 3 3	1 3 1 4 4	2 10 6 4 6 4 6	26 25 10 11 19 17	20 12 20 22 12 19 15	1 1 2 1 2 3 2	1				
GOC-1 GOC-3 GOC-10 GOC-11		3	2 1 1	2 1 5 2	6 6 11 4	17 21 24 7	20 17 9 18	3 1 17	1				
GOF-1 GOF-3 GOF-4 GOF-5 GOF-6 GOF-7 GOF-11 GOF-13	4 2 4 4	2 5 3 1	5 1 2 1 1 4	3 4 1 4 2 2 3	13 3 17 13 17 22 4 19	27 22 24 23 11 16 26 19	9 11 2 5 9 2 17	1 1 6	1				
GOX-1 GOX-2 GOX-3 GOX-4 GOX-5 GOX-6			2 1 1	3 2 2 2 6 1	10 10 2 5 10 10	18 21 20 22 22 22 20	19 14 15 17 12 19	1 7 3	3				
LA-1 LA-3 LA-4	2 2 3	1 1 4	2 2 5	1 2 5	4 1 5	13 15 7	14 19 17	6 5 3	5 2 1	2 1			
LC-5 LC-6 LC-8 LC-9	2 2	2 3 3 3	2 3 5 3	5 5 6 4	4 7 7 4	10 13 9 11	13 11 10 15	3 2 4 4	4 2 4 3	3 1 1 1	1 1 1 1	1	
LE-3 LE-5	1	1	2	1 3	5 11	18 25	17 11	3	2				
LH-1 LH-3	3	5 3	7 5	5 5	6 8	13 18	10 10	1					

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### THE THELEPHORACEAE OF NORTH AMERICA. XV1

(Conclusion, With Supplement and General Index)

#### EDWARD ANGUS BURT

Mycologist and Librarian to the Missouri Botanical Garden

#### CORTICIUM

Corticium Persoon, Roemer Neues Mag. Bot. 1: 110. 1794; Obs. Myc. 1: 37. 1796; Fries, Gen. Hym. 15. 1836; Epicr. 1838; Hym. Eur. 646. 1874; Berkeley, Outl. Brit. Fung. 1860; Morgan, Cincinnati Soc. Nat. Hist. Jour. 10: 198. 1888; Sacc. Syll. Fung. 6: 603. 1888; Karsten, Vet.-Soc. Bidrag Natur och Folk 48: 408. 1889; Massee, Linn. Soc. Bot. Jour. 27: 117. 1890; Bresadola, I. R. Accad. Agiati Atti III. 2: 110. 1897; Ann. Myc. 1:93. 1903; Engl. & Prantl, Nat. Pflanzenfam. (I: 1\*\*): 118. 1898; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 1911; Rea, Brit. Basid. 14, 672. 1922.—Includes Gloeocystidium v. Höhnel & Litschauer, Weisner Festschr. Wien, 58. 1908, and Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 354. 1913. -Not Gloeocystidium Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 48: 429. 1889. See Burt, Mo. Bot. Gard. Ann. 12: 247. 1926.—Includes Vararia Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 52: 96. 1898; Asterostromella v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 773. 1907; Weisner Festschr. Wien, 58. 1908; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 265. 1911.—Includes Xerocarpus and Lyomyces of Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 48: 417, 418. Includes in part Hypochnus Sacc. Syll. Fung. 6: 653. 1888, and Engl. & Prantl, Nat. Pflanzenfam. (I: 1\*\*): 116. 1898.—Not Hypochnus Fries emend. Karsten, Rev. Myc. 3º: 23. 1881. See Burt, Mo. Bot. Gard. Ann. 3: 203.

<sup>&</sup>lt;sup>1</sup> Issued September 20, 1926.

Fructifications waxy, crustaceous or floccose, fleshy, cartilaginous, coriaceous or membranaceous, always resupinate, effused; hymenium even, or somewhat tubercular in a few species; basidia simple with 2-8 sterigmata, usually 4, the sterigmata not greatly thickened; basidiospores white, even—green in C. atrovirens; substance variously differentiated but not containing colored, stellate organs. Distinguished from Peniophora by not having cystidia.

The species described as belonging in Corticium upon publication of the genus are Corticium polygonium, C. laeve, C. roseum, C. Sambuci, C. cinereum, and C. aurantium, in the order given, no one of which was designated as the type species. C. Sambuci and C. cinereum are now included in Peniophora and C. aurantium in Aleurodiscus.

Von Höhnel and Litschauer and Bourdot and Galzin have segregated under Gloeocystidium Karsten all species of Corticium which have gloeocystidia. I have not followed them in this, because I regard gloeocystidia as but one of the several differentiations of tissue which afford helpful distinctive characters for recognition of the species of this genus. In fact, I feel that closer observation of the tissues and structure of the fructification and accurate record of such structure should give important, and often decisive, characters of all the species. My own study has already gone so far in this direction that I attach but slight regard to a specific determination which is based merely upon obvious external characters and the substratum upon which growing. A sufficient objection to Gloeocystidium for the species which have gloeocystidia is that one of the two species upon which Karsten founded the genus is Peniophora guttulifera, a true Peniophora with no gloeocystidia whatever, and the other is Odontia sudans.

Asterostromella as a genus to include Corticium investiens, a species with helpful hyphal differentiation, is antedated by Karsten's Vararia, having C. investiens as its type species.

What was shown in the preceding part about the distribution of our species of *Peniophora* is true also for *Corticium*. Of the 107 species of *Corticium* herein presented, 46 are described as new species. The color of the exterior of the fructification and of its internal substance and the presence of tissues of somewhat unusual form have afforded a basis for the arrangement of our

species into 4 workable groups nearly equal in numbers, as presented in the following key to the species. Each of these groups is subdivided to such degree as seems desirable—largely by spore characters—into minor groups of so few species that the characters of the component species of any group may, and should, all be considered in determining the probable species of the specimen in course of identification. The extensive lists of specimens studied, with their localities where collected, and present preservation in published exsiccati and herbaria, afford material for checking up one's own determinations. Determinations as published should correct earlier tentative determinations communicated to my correspondents.

Throughout this work technical color terms are those of Ridgway's 'Color Standards and Nomenclature.' There was little knowledge available as to the color of specimens when growing, but since the time-consuming task of determination is usually with dried specimens collected many years ago and often more or less faded or yellowed, my record of the color of the dried specimens should be the more helpful to the chief users of this work.

Accounts of the species of the genera Tremellodendron, Eichleriella, Sebacina and Septobasidium were included to set off more sharply the true Thelephoraceae to which the species of these four genera are so similar in aspect that they were commonly known under their original names as species of Thelephoraceae. By treating these genera and Lachnocladium in the present work, the student had at hand a systematic account of all North American fungi of thelephoraceous aspect. The matter on those genera could otherwise have been included in my recent publications: 'Some North American Tremellaceae, Dacryomycetaceae and Auriculariaceae' and 'North American species of Clavaria.'

To all whose names have been recorded as collectors and contributors of specimens and to botanical institutions whose specimens are cited and which have afforded me facilities for the study of their herbaria I am deeply indebted. Without their aid but little could have been done.

# KEY TO THE SPECIES

I.	Substance not appreciably colored, no gloeocystidia.	
	1. Hymenium white or whitish when growing	1-23
	*With antier-shaped paraphyses or color change from yellow to	
	white in fruiting	1, 2
	**Spores globose or subglobose.	,
	a. Imbedded spores (chlamydospores) usually present	3, 4
	b. Imbedded spores not yet observed	5-9
	***Spores more elongated.	00
	a. Spores large, more than 6 $\mu$ long	11 99
	b. Spores small, hyphae incrusted or among obscuring mineral	11,00
	matter	1 <b>2</b> –16
	~	15-23
		10-23
	2. Hymenium colored when dry and not known to be white at first—	01 50
	usually some shade of buff, yellow, red, brown or blue	24-56
	*Spores globose or subglobose, less than 5 $\mu$ in diameter	24-26
	**Spores globose or subglobose, more than 5 $\mu$ in diameter	<b>2</b> 7- <b>2</b> 9
	***Spores more elongated.	
		30, 31
	b. Spores large, 6–12 $\mu$ long	
	c. Spores small, hyphae somewhat incrusted	
	d. Spores small, hyphae not incrusted, fructifications separable.	49
	e. Spores small, hyphae not incrusted, fructifications closely	
	adnate or only small pieces separable	<i>50–56</i>
II.	Gloeocystidia present or structure vesicular, or some tissue note-	
	worthy, substance colored or not colored.	
	*Gloeocystidia present or shown by vesicular structure or by colored,	
	resinous-appearing masses	36, 107
	a. Gloeocystidia not colored, elongated, imbedded spores nu-	
	merous	<i>57-58</i>
	b. Gloeocystidia not colored, elongated, lacking chlamydospores.	
	†Spores globose, subglobose or broadly ovoid	<i>59-66</i>
	††Spores more elongated	71. 79
	c. Gloeocystidia not colored, pyriform to globose	72-79
	d. Gloeocystidia colored, elongated	80-83
	e. Gloeocystidia colored, subangular or globose, resinous-ap-	
	pearing	84-86
	**Distinguished by antler-shaped branching of some hyphae or para-	04 00
	physes, or other branching of paraphyses, or unusual form of other	
	tissues	1 107
	**Numerous imbedded spores or other than basidiospores3, 4, 11, 37,	
	***Spores green, even	105
		34
	**Spores usually white but finally becoming ochraceous	34 37-106
III		,,-100
	*Fructifications ranging from gray to drab.	07 00
	a. With paraphyses having slender branches, spores small	87, 88 89-91
	b. Paraphyses not noteworthy, spores larger, 7-10 μ long	08-81
	**Fructifications ochraceous to wax-yellow and red.	

a.	With some hyphae or paraphyses having antier-shaped or	
	racemose branching	92-94
b.	Tissues not having antler-shaped or racemose branching	2, 95-99
***Fructif	ications darker, tending to brown and vinaceous.	
a.	Parasitic species	100-102
b.	Always saprophytic	103, 104
****Fructif	ications green or blue	105, 106

## 1. Corticium paraphysatum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, thin, closely adnate, white to pale cartridge-buff in the herbarium, even, velutinous, not shining, not cracked, the margin similar, thinning out; in section 45–75  $\mu$  thick, not colored, composed of somewhat scattered, deeply staining, clavate organs—probably basidia—immersed among great numbers of slender, erect, non-staining, branching organs which approach antler-form in branching and form the layer of paraphyses at the surface of the hymenium; no gloeocystidia; no basidia bearing sterigmata nor spores found.

Fructifications 1-5 cm. long,  $\frac{1}{2}$ -2 $\frac{1}{2}$  cm. wide. Small fructifications become confluent.

Beneath prostrate, decaying, hardwood limbs of a frondose species. Cuba. Still immature in December.

Although the specimens at hand of *C. paraphysalum* are still so immature that it has not been possible to demonstrate their mature basidia and spores, the species is distinct from others of the genera *Aleurodiscus*, *Sebacina*, and *Corticium* which are known to me. It should be readily recognizable by its thin, closely adnate, white fructifications on small hardwood limbs and by the abundance of the non-staining paraphyses.

Specimens examined:

Cuba: Ceballos, C. J. Humphrey, 2848, type, and 2776, 2800 (in Mo. Bot. Gard. Herb., 63769, 63768, and 63770 respectively), and 2586; Omaja, C. J. Humphrey, 2698 (in Mo. Bot. Gard. Herb., 43063).

C. sulphureum Fries, Epicr. 561. 1838; Hym. Eur. 650.
 1874; Berkeley, Outl. Brit. Fung. 274. 1860; Sacc. Syll. Fung.
 6: 612. 1888.

Thelephora sulphurea Fries, Syst. Mcy. 1:452. 1821; Elenchus

Fung. 1: 204. 1828.—Corticium croceum Bresadola, I. R. Accad. Agiati Atti III. 3: 112. 1897; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 242. 1911; Rea, Brit. Basid. 676. 1922.—An Sporotrichum croceum Kunze & Schmidt, Myk. Heft. 1: 81. 1817?—Not Corticium sulphureum Persoon, which is a synonym of Hypochnus fumosus Fries. See Burt, Mo. Bot. Gard. Ann. 3: 239. 1916.

Type: authentic specimen in Kew Herb.

Fructifications effused, fibrillose-byssoid, sulphur-yellow to wax-yellow when a sterile mycelium, becoming whitish throughout when forming the hymenium, the margin yellow or whitish, running out into sulphur-yellow to wax-yellow branching rhizomorphic strands; when fertile  $200-300\,\mu$  thick in section, not appreciably colored, the hyphae loosely arranged, ascending, branching,  $2\frac{1}{2}\,\mu$  in diameter, rough-walled or somewhat incrusted with small crystals; no gloeocystidia; spores hyaline, even,  $3\times 2\,\mu$ , copious.

Fructifications 3-10 cm. long, 2-4 cm. wide.

Under side of decaying Fagus and other species. Europe, Maryland, Missouri, Montana, and Idaho. Common in Europe but rare in North America. August to October.

The mycelium of *C. sulphureum* is conspicuous by its brilliant wax-yellow color, but in fruiting this yellow color is lost throughout the fructification, persisting only about the margin and in the rhizomorphic strands. By this curious character and by the pruinose or velvety hymenium one may distinguish *C. sulphureum* from *C. bicolor*. The International Botanical Rules afford no ground for the displacement by Bresadola of the well-established name *Corticium sulphureum* by *C. croceum*.

Specimens examined:

Sweden: authentic specimen from E. Fries (in Kew Herb.); Femsjö, E. A. Burt, 2 gatherings; Stockholm, L. Romell, 151, 152.

Germany: Brinkmann, comm. by G. Bresadola.

Austria: Innsbruck, V. Litschauer; Tirol, V. Litschauer.

Maryland: Takoma Park, C. L. Shear, 1069.

Missouri: Meramec Highlands, F. P. McWhorter (in Mo. Bot. Gard. Herb., 57359).

Montana: Bernice, E. E. Hubert, comm. by J. R. Weir, 12008 (in Mo. Bot. Gard. Herb., 63368).

Idaho: Priest River, E. E. Hubert, comm. by J. R. Weir, 12021 (in Mo. Bot. Gard. Herb., 63376).

3. C. punctulatum Cooke, Grevillea 6: 132. 1878; Sacc. Syll. Fung. 6: 614. 1888; Massee, Linn. Soc. Bot. Jour. 27: 129. 1890.

Type: type distribution in Ravenel, Fungi Am., 128.

Fructifications broadly effused, thin, somewhat hypochnoid, only fragments separable, white at first, becoming between pinkish buff and cream-color in the herbarium, punctulate at first, at length even and continuous in spots, fibrillose, the margin thinning out, concolorous, indeterminate; in section about 135  $\mu$  thick, not colored, with hyphae loosely interwoven,  $4-4\frac{1}{2}\mu$  in diameter, not incrusted, occasionally nodose-septate; no gloeocystidia; spores imbedded in all regions of the fructification are probably chlamydospores; basidia bearing sterigmata or spores not demonstrated; spores at surface of hymenium hyaline, even, perhaps becoming minutely rough,  $6 \times 4\frac{1}{2}-5\mu$ , copious.

Fructifications up to 6 cm. long, 1-2 cm. wide.

On rotten pine logs and on small splinters and rubbish consolidated by the mycelium. New Jersey and South Carolina.

The punctulate hymenium of *C. punctulatum* is distinctive in the several specimens from the original collection now in three herbaria; the presence of imbedded spores in all regions of the fructification should prove another helpful character for the recognition of this species.

Specimens examined:

Exsiccati: Ravenel, Fungi Am., 128.

New Jersey: Belleplain, C. L. Shear, 1248.

South Carolina: Aiken, H. W. Ravenel, 2334, type (in Kew Herb. and in Ravenel, Fungi Am., 128).

4. C. vellereum Ellis & Cragin, Jour. Myc. 1: 58. 1885; Sacc. Syll. Fung. 6: 615. 1888; Massee, Linn. Soc. Bot. Jour. 27: 137. 1890; Wakefield, Brit. Myc. Soc. Trans. 5: 128. 1914.

Corticium Bresadolae Bourdot, Rev. Sci. Bourb. 23: 6. 1910; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 233. 1911. Type: in N. Y. Bot. Gard. Herb.

Fructifications widely effused, adnate, rather thick, tender, small pieces separable when moistened, white, cream-buff or pinkish buff, even, pulverulent or waxy, rarely cracked, the margin white, byssoid; in section 200–500  $\mu$  thick, not colored, composed of loosely interwoven, thin-walled, nodose-septate hyphae 3–5  $\mu$  in diameter and usually numerous chlamydospores; no gloeocystidia; basidiospores white in spore collection, even, subglobose, 5–7  $\times$  4½–6  $\mu$ ; chlamydospores of about the same dimensions.

Fructifications 3-10 cm. in diameter.

On bark and wood of frondose species decaying on the ground. In Europe, from Canada to Texas, westward to British Columbia and California, and in Mexico and Japan. July to March. Common.

C. vellereum is distinguished among our species of Corticium by the presence usually of very numerous chlamydospores and by the absence of gloeocystidia. This is true of C. punctulatum, but the latter is more hypochnoid in surface and occurs on pine.

Specimens examined:

Sweden: L. Romell, 404.

France: St. Priest, H. Bourdot, 15749, authentic specimen of C. Bresadolae.

England: Winchester, F. Escombe, comm. by E. M. Wakefield (in Mo. Bot. Gard. Herb., 4038).

Canada: J. Macoun, 652, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 7457); Ottawa, J. Macoun, 8, 43, 180, and 281 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57455); St. Lawrence Valley, J. Macoun, 25.

New Hampshire: Chocorua, E. A. Burt.

Vermont: Middlebury, E. A. Burt; Abby Pond, Ripton, E. A. Burt.

Massachusetts: Magnolia, W. G. Farlow.

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 59689); Hudson Falls, S. H. Burnham, 13 (in Mo. Bot. Gard. Herb., 44004); Ithaca, G. F. Atkinson, 22971; Jordan, E. Brown, 179 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61451); Van Cortland Park, New York

- City, W. A. Murrill (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61429); Westport, C. H. Peck, 2 (in N. Y. State Mus. Herb., T 24, and Mo. Bot. Gard. Herb., 56070).
- Pennsylvania: State College, L. O. Overholts, 4811 (in Mo. Bot. Gard. Herb., 56125).
- Georgia: Savannah, C. J. Humphrey, 5109 (in Mo. Bot. Gard. Herb., 11953).
- Alabama: Auburn, F. S. Earle, 115 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61561).
- Texas: Quitman, W. H. Long, comm. by C. J. Humphrey, 2545 (in Mo. Bot. Gard. Herb., 9920).
- Ohio: C. G. Lloyd, 3738, 3825; Linwood, C. G. Lloyd, 1880.
- Michigan: Ann Arbor, C. H. Kauffman, 11, 16.
- Wisconsin: Superior, C. J. Humphrey, 1548 (in Mo. Bot. Gard. Herb., 10744).
- Illinois: River Forest, E. T. & S. A. Harper, 627, 629.
- Missouri: Upper Creve Coeur, E. A. Burt (in Mo. Bot. Gard. Herb., 58345); St. Louis, S. M. Zeller (in Mo. Bot. Gard. Herb., 55642); Valley Park, E. A. Burt (in Mo. Bot. Gard. Herb., 44074).
- Kansas: Rooks County, E. Bartholomew, 2 specimens under the herbarium name C. globiferum (in Burt Herb., and Mo. Bot. Gard. Herb., 4848, 4849); Strong City, G. G. Hedgcock, comm. by C. J. Humphrey, 2541 (in Mo. Bot. Gard. Herb., 11043); Topeka, F. W. Cragin, 560, type, 583 (in N. Y. Bot. Gard. Herb.).
- South Dakota: Black Hills, J. R. Weir, 10014 (in Mo. Bot. Gard. Herb., 55795).
- Idaho: Priest River, E. E. Hubert, comm. by J. R. Weir, 11633 (in Mo. Bot. Gard. Herb., 63306).
- Manitoba: Winnipeg, A. H. R. Buller, 720, 845 (in Mo. Bot. Gard. Herb., 58984, 58993); G. R. Bisby, 1341, 1347 (in Mo. Bot. Gard. Herb., 60550, 60557).
- British Columbia: G. M. Dawson, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 44690).
- California: Berkeley, W. T. Horne, comm. by W. A. Setchell, 1031 (in Mo. Bot. Gard. Herb., 44239).
- Mexico: Guernavaca, W. A. & E. L. Murrill, 361, 371, comm. by

N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54464, 54459); Parral, Chihuahua, E. O. Mathews, 19 (in Mo. Bot. Gard. Herb., 44127).

Japan: Kogura Prov., Kozuka, A. Yasuda, 154 (in Mo. Bot. Gard. Herb., 62956).

5. C. granulare Burt, Mo. Bot. Gard. Ann. 10: 187. 1923. Type: in Mo. Bot. Gard. Herb.

Fructification effused, adnate, snow-white, pulverulent under a lens, very thin, only 15–30  $\mu$  thick, not bearing a continuous hymenium but consisting of bushy branched, suberect hyphal clusters standing out from the substratum and near together, with their main trunks up to 6  $\mu$  in diameter and short-celled; no cyrtidia nor gloeocystidia; basidia simple,  $15 \times 4\frac{1}{2} \mu$ , with 4 sterigmata; spores hyaline, even, flattened on one side,  $4-4\frac{1}{2} \times 3-4 \mu$ , copious.

Fructifications scattered along the substratum, 1-3 cm. long, 4-8 mm. wide.

On dead herbaceous stems. Hawaiian Islands, F. L. Stevens, 381, type (in Mo. Bot. Gard. Herb., 60603).

# 6. C. ermineum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, thin, closely adnate, white, not shining, not cracked, the margin similar, thinning out, fimbriate; in section 200  $\mu$  thick, not colored, with some hyphae densely arranged along the substratum but becoming suberect and more loosely arranged towards the hymenium, 3  $\mu$  in diameter, incrusted, not nodose-septate; no gloeocystidia; spores hyaline, even,  $7-9 \times 5-6 \mu$ , copious.

Fructifications up to 12 cm. long, 3 cm. wide.

On decorticated, very rotten wood of logs of *Thuja plicata* and spruce. Vermont and Idaho. August and October.

C. ermineum is distinct among our white species of Corticium by its ermine-white color, well-incrusted hyphae, large spores and occurrence on coniferous wood. C. amylaceum of France, of which I have a cotype, is a related species but thinner, more farinose, and less compact.

Specimens examined:

Vermont: Middlebury, E. A. Burt.

Idaho: Priest River, E. E. Hubert, comm. by J. R. Weir, 12026, type (in Mo. Bot. Gard. Herb., 63379).

C. Berkeleyi Cooke in Massee, Linn. Soc. Bot. Jour. 27:
 133. 1890; Sacc. Syll. Fung. 11: 127. 1895.

Type: type distribution in Ravenel, Fungi Am., 225.

Fructifications broadly effused, thin, membranaceous-arachnoid, small pieces separable when moistened, whitish at first, becoming light buff to pinkish buff in the herbarium, even or minutely granular, not waxy nor shining, cracked, the margin thinning out, with hyphae interwoven; in section 100–200  $\mu$  thick, not colored, with hyphae nodose-septate, not incrusted, 4–5  $\mu$  in diameter along the substratum and sending out ascending, loosely arranged branches which become smaller and densely arranged in the hymenium; no gloeocystidia; basidia 4-spored; spores hyaline, even, subglobose and 4–8  $\times$  4–6  $\mu$ , or globose and 4–6  $\mu$  in diameter.

Fructifications 3-10 cm. in diameter.

On bark and wood of conifers—usually pine. Canada to Texas and in Michigan, Idaho, British Columbia, and New Mexico. April to November. Infrequent.

C. Berkeleyi probably covers large areas on bark of pine logs. It is white or very nearly white, with the hymenium barely continuous, spores globose or subglobose, and hyphae coarse and mostly erect, like those of C. bombycinum but with not as thick fructifications and a very inconspicuous margin in comparison with C. bombycinum.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 934; Ravenel, Fungi Am., 225, type distribution.

Canada: J. Macoun, 32; Lower St. Lawrence Valley, J. Macoun, 74.

Ontario: Ottawa, J. Macoun, 35.

New Hampshire: Chocorua, W. G. Farlow, 9.

Vermont: Middlebury, E. A. Burt.

New York: Newtonville, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 14854).

North Carolina: Chapel Hill, J. N. Couch, comm. by W. C. Coker, 4257 (in Mo. Bot. Gard. Herb., 57419).

South Carolina: Aiken, H. W. Ravenel, in Ellis, N. Am. Fungi, 934.

Georgia: Darien, H. W. Ravenel, in Ravenel, Fungi Am., 225; Savannah, C. J. Humphrey, 5109 (in Mo. Bot. Gard. Herb., 11953).

Alabama: Montgomery County, R. P. Burke, 519 (in Mo. Bot. Gard. Herb., 57305).

Texas: Quitman, W. H. Long, comm. by C. J. Humphrey, 2545 (in Mo. Bot. Gard. Herb., 9920).

Michigan: Ann Arbor, C. H. Kauffman, 34.

Idaho: Kooskia, J. R. Weir, 397 (in Mo. Bot. Gard. Herb., 13544); Priest River, J. R. Weir, 6360 (in Mo. Bot. Gard. Herb., 58449).

British Columbia: Kootenai Mts., near Salmo, J. R. Weir, 478 (in Mo. Bot. Gard. Herb., 63274).

New Mexico: Cloudcroft, W. H. Long, 19523 (in Mo. Bot. Gard. Herb., 44767); Mogollen, G. G. Hedgcock & W. H. Long, comm. by C. J. Humphrey, 2559 (in Mo. Bot. Gard. Herb., 9781).

8. C. arachnoideum Berkeley, Ann. & Mag. Nat. Hist. 13: 345. pl. 9, f. 3. 1844; Outl. Brit. Fung. 273. 1860; Berk. & Curtis, Grevillea 2: 4. 1873; Fries, Hym. Eur. 649. 1874; Sacc. Syll. Fung. 6: 611. 1888; Massee, Linn. Soc. Bot. Jour. 27: 135. 1890; Bresadola, Ann. Myc. 1: 93. 1903.

Not probably C. arachnoideum as understood by v. Höhnel & Litschauer, and Rea.

Type: in Kew Herb.

Fructifications effused, thin, arachnoid, tender, snow-white, forming an even hymenial pellicle in the older, more central portions, supported on the loosely arranged arachnoid subiculum which protrudes as a sterile, delicate, web-like margin; in section  $100-200 \mu$  thick, not colored, with hyphae very loosely interwoven,  $3-4 \mu$  in diameter, nodose-septate, not incrusted; no gloeocystidia; spores hyaline, even, globose, or subglobose,  $4-6 \mu$  in diameter or  $6 \times 5 \mu$ ,  $5 \times 4 \mu$ ,  $4-4\frac{1}{2} \times 3-4 \mu$ .

Fructifications 2-6 cm. long, 1-3 cm. wide.

On humus of leaf fragments and decaying wood, running over

mosses and lichens and on rotten wood. Rare in Europe, common in North America from Canada to Louisiana and westward to the Pacific, in the West Indies and the Hawaiian Islands. May to November.

C. arachnoideum is globose-spored and separated from C. lacteum by white color, more arachnoid subiculum, and thinner and less compact hymenium. C. centrifugum, which is common in Europe and infrequent in North America, has narrower spores than C. arachnoideum, is less arachnoid, more inclined to ashy white color, more widely effused, and on decaying wood preferably. Our American specimens of C. arachnoideum agree perfectly with those of Berkeley in Kew and with the Berkeley & Curtis specimens also determined by Berkeley.

Specimens examined:

Exsiccati: Brinkmann, Westfälische Pilze, 103; Ellis, N. Am. Fungi, 411; Ell. & Ev., Fungi Col., 918.

Sweden: L. Romell, 77; Stockholm, L. Romell, 161.

England: on moss, 437, authentic specimen, perhaps type, M.J. Berkeley (in Kew Herb.).

Scotland: Glamis, J. Stevenson (in Berkeley Herb. of Kew Herb.).

Germany: Westphalia, W. Brinkmann, comm. by Bresadola, and in Brinkmann, Westfälische Pilze, 103 (in Mo. Bot. Gard. Herb., 63441).

Austria: Stubai, Tirol, V. Litschauer, under the name Corticium centrifugum var. macrosporum.

Canada: J. Macoun, 47, 63; Lower St. Lawrence Valley, J. Macoun, 12, 64, 89; London, Ontario, J. Dearness, 1146 (in Mo. Bot. Gard. Herb., 18762); Ottawa, J. Macoun, 400.

Newfoundland: Bay of Islands, A. C. Waghorne, 1014 (in Mo. Bot. Gard. Herb., 4813).

Massachusetts: Sharon, A. P. D. Piguet, comm. by W. G. Farlow, and 135, comm. by Farlow Herb. (in Mo. Bot. Gard. Herb., 59626).

Vermont: Middlebury, E. A. Burt, 4 gatherings.

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57509); Bolton, C. H. Peck, 17; Bolton Landing, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55769); East Galway, E. A. Burt; Ithaca, G. F.

- Atkinson, 2125, 8054, 8240, 14356; H. S. Jackson, 18658; C. Thom, 14367; Karner, H. D. House, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 55193).
- New Jersey: Newfield, J. B. Ellis, in Ellis, N. Am. Fungi, 411, Ell. & Ev., Fungi Col., 918, and 1374, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 14652).
- Maryland: Takoma Park, C. L. Shear, 1029, 1105.
- North Carolina: Blowing Rock, G. F. Atkinson, 4325; Chapel Hill, J. N. Couch, comm. by W. C. Coker, 4235a (in Mo. Bot. Gard. Herb., 57418).
- South Carolina: M. A. Curtis, 2513 (in Farlow Herb.).
- Mississippi: Ocean Springs, L. M. Underwood (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61482).
- Louisiana: Plaqueminas County, A. B. Langlois, 998.
- Illinois: Riverside, E. T. & S. A. Harper, 738.
- Montana: Hecla, E. E. Hubert, comm. by J. R. Weir, 11408 (in Mo. Bot. Gard. Herb., 63264); Missoula, J. R. Weir, 402 (in Mo. Bot. Gard. Herb., 11256); Rock Hill, J. R. Weir, 11963 (in Mo. Bot. Gard. Herb., 63224); Yellow Bay, J. A. Hughes, comm. by J. R. Weir, 7035 (in Mo. Bot. Gard. Herb., 55466).
- Idaho: Coolin, J. R. Weir, 11540 (in Mo. Bot. Gard. Herb., 63295); Ruby Creek, E. E. Hubert, comm. by J. R. Weir, 12009 (in Mo. Bot. Gard. Herb., 63369); Sandpoint, E. E. Hubert, comm. by J. R. Weir, 12024 (in Mo. Bot. Gard. Herb., 63377).
- Manitoba: Norway House, G. R. Bisby, 1465 (in Mo. Bot. Gard. Herb., 57912).
- Washington: Falcon Valley, W. N. Suksdorf, 725; Mt. Paddo, W. N. Suksdorf, 734; Sedro-Woolley, C. J. Humphrey, 1045 (in Mo. Bot. Gard. Herb., 10901).
- Oregon: Wallowa Lake, C. L. Shear, 798.
- California: Redding, C. J. Humphrey, 1045; Santa Catalina Island, L. W. Nuttall, 1092 (in Mo. Bot. Gard. Herb., 58871).
- Cuba: San Diego de los Baños, Earle & Murrill, 361, comm. by N. Y. Bot. Gard. Herb.
- Porto Rico: Rio Piedras, J. A. Stevenson, 6557 (in Mo. Bot. Gard. Herb., 55080).
- Hawaiian Islands: F. L. Stevens, 964 (in Stevens Herb., Mo. Bot. Gard. Herb., 60602, and Burt Herb.).

C. portentosum Berk. & Curtis, Grevillea 2: 3. 1873;
 Morgan, Cincinnati Soc. Nat. Hist. Jour. 10: 201. 1888;
 Sacc. Syll. Fung. 6: 636. 1888;
 Massee, Linn. Soc. Bot. Jour. 27: 129. 1890;
 Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 235. 1911.

Corticium diminuens Berk. & Curtis, Grevillea 2: 3. 1873; Sacc. Syll. Fung. 6: 631. 1888; Massee, Linn. Soc. Bot. Jour. 27: 158. 1890.—Stereum portentosum (Berk. & Curtis) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 743. 1907.—Corticium portentosum crystallophorum Ell. & Ev. Torr. Bot. Club Bul. 24: 125. 1897.—Corticium Aluta Bresadola in v. Höhnel & Litschauer, Wiesner Festschr. Wien, 62. 1908.—An Corticium grammicum P. Hennings, Engl. Bot. Jahrb. 38: 106. 1905? Compare v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 743. 1907.

Type: in Kew Herb. and Curtis Herb.

Fructifications long and widely effused, thick, coriaceous-soft, small pieces separable when moistened, white, becoming light buff to warm buff in the herbarium, even, only rarely cracked, the margin often whitish, pubescent-villose; in section 150–1000  $\mu$  thick, colored like the hymenium, becoming zonate or stratose when thick, composed of very densely interwoven, tough hyphae about 1–2  $\mu$  in diameter, not incrusted, not nodose-septate, protruding in the hymenial surface as curved paraphyses; more or less numerous aggregations of mineral matter may be immersed in the substance; no gloeocystidia; basidia few; spores hyaline, even, spherical,  $4\frac{1}{2}$ –7  $\mu$  in diameter, few present usually.

Fructifications 4-12 cm. long, 2-4 cm. wide.

On bark and wood of logs of frondose species. In Europe, South Africa, throughout North America and the West Indies, in South America, and in the Philippine Islands. Common.

C. portentosum is well named and may be recognized by its large, whitish, coriaceous fructifications on frondose logs, which become zonate within in thick specimens, and have globose spores  $6 \mu$  in diameter, and the slender branches of the interwoven hyphae exceeding the basidia and forming the hymenial surface. This species was formerly confused in Europe with Stereum alneum and was communicated to me under this name by both Karsten and Bresadola. It also occurs from Lindblad in Kew

Herb. and from Blytt in Fries Herb. under the name of *Stereum odoratum*, from another specimen of which, determined by E. Fries, it differs by the elongated spores and occurrence on *Pinus* of the latter.

Specimens examined:

- Exsiccati: Ellis, N. Am. Fungi, 718; Ell. & Ev., N. Am. Fungi, 1715; Ravenel, Fungi Car. 3: 31; de Thuemen, Myc. Univ., 2013, under the name *Corticium radiosum*.
- Finland: Mustiala, P. A. Karsten, in de Thuemen, Myc. Univ., 2013; Vasa, P. A. Karsten, under the name Stereum alneum.
- Sweden: Stockholm, L. Romell, 26, 159, both under the name Stereum alneum.
- Germany: Feldkirch, Rick, comm. by Bresadola, under the name Stereum alneum.
- Hungary: Kmet, comm. by Bresadola, under the name Stereum odoratum.
- Italy: locality not stated, Bresadola, comm. under the name Stereum alneum; Trento, Bresadola.
- France: Aveyron, A. Galzin, 14990, comm. by H. Bourdot, 15750. Canada: Ontario, London, J. Dearness, 1287 (in Mo. Bot. Gard. Herb., 19057).
- New York: Ithaca, G. F. Atkinson, 3406; Poughkeepsie, W. R. Gerard, 316 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61385).
- Pennsylvania: Michener, type (in Kew Herb., and Curtis Herb., 3620); West Chester, Everhart, Haines, Jefferis & Gray, in Ellis, N. Am. Fungi, 718.
- Florida: W. W. Calkins, in Ell. & Ev. N. Am. Fungi, 718, and (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 61488, and Burt Herb.); H. von Schrenk (in Mo. Bot. Gard. Herb., 44202); Cocoanut Grove, R. Thaxter, 99 (in Mo. Bot. Gard. Herb., 43926); Cutler Hammock, W. A. Murrill, 76, 252, 253, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 62101, 62129, and 62128, respectively); Miami, W. H. Long, 12290 (in Mo. Bot. Gard. Herb., 55051); Palm Beach, R. Thaxter, 15 (in Mo. Bot. Gard. Herb., 43928).
- Alabama: Peters, type distribution of Corticium diminuens, in Ravenel, Fungi Car. 3:31, and (in Curtis Herb., 4009); Mont-

- gomery County, R. P. Burke, 464 (in Mo. Bot. Gard. Herb., 57285).
- Louisiana: A. B. Langlois, 244, comm. by U. S. Dept. Agr. Herb.; St. Martinville, A. B. Langlois, 1762, 2098, and 1247, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 44075), and 2438, type of Corticium portentosum crystallophorum.
- Texas: San Antonio, W. H. Long, 21187 (in Mo. Bot. Gard. Herb., 55132); Uvalde, W. H. Long, 21686 (in Mo. Bot. Gard. Herb., 55133).
- Kentucky: Mammoth Cave, C. G. Lloyd, 2568.
- Ohio: Cincinnati, A. P. Morgan (in Lloyd Herb., 2604, and under the name Corticium subgiganteum); Loveland, D. L. James (in U. S. Dept. Agr. Herb.); West Elkton, L. O. Overholts, 4208 (in Mo. Bot. Gard. Herb., 55637); Waynesville, F. G. Lea, the C. ochraceum of Lea's Cat. Plants of Ohio (in Berkeley Herb. at Kew).
- Indiana: Scottsburg, J. R. Weir, 369 (in Mo. Bot. Gard. Herb., 17771); Weirtown, J. R. Weir, 353 (in Mo. Bot. Gard. Herb., 9933).
- Wisconsin: Lake Geneva, E. T. & S. A. Harper, 848; Star Lake, Miss Stucki, 56.
- Missouri: Columbia, B. M. Duggar, 569.
- British Columbia: Sidney, J. Macoun, 24, 37, 86, 88, 105, 165 (in Mo. Bot. Gard. Herb., 5685, 55348, 8109, 11350, 55349, 20477);
  Squamish, J. Macoun, 537, 570 (in Mo. Bot. Gard. Herb., 55192, 55185);
  Vancouver Island, J. Macoun, 144, 295, 537 (in Mo. Bot. Gard. Herb., 18865, 55320, 55319).
- Mexico: Jalapa, W. A. & E. L. Murrill, 115, 191, 346, comm. by
  N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 10854, 54437, 54481); Orizaba, W. A. & E. L. Murrill, 750, comm. by
  N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54636).
- Bermuda: Paget Swamp, H. H. Whetzel, Abf (in Mo. Bot. Gard. Herb., 58910).
- Cuba: Baracoa, L. M. Underwood & F. S. Earle, 784, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 61556); Camaguey (in Mo. Bot. Gard. Herb., 56123); Havana Province, Earle & Murrill, 24, 103, comm. by N. Y. Bot. Gard. Herb.; Omaja, C. J. Humphrey, 2709, 2830 (in Mo. Bot. Gard. Herb.,

13740, 14847); Oriente, comm. by J. R. Weir 10617 (in Mo. Bot. Gard. Herb., 56235); Pinar del Rio Province, Earle & Murrill, 196, 201, 208, 295, 312, comm. by N. Y. Bot. Gard. Herb., P. Wilson, 11570, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 61494); Puerto Principe Province, Earle & Murrill, 582, 602, comm. by N. Y. Bot. Gard. Herb.; Santiago de Cuba Province, Earle & Murrill, 460, 467, comm. by N. Y. Bot. Gard. Herb.

Porto Rico: Ponce, F. S. Earle, 117; Rio Piedras, J. R. Johnston, 982, 982a, 972a (in Mo. Bot. Gard. Herb., 9849, 61355, 61356), J. A. Stevenson, 3597, 5158 (in Mo. Bot. Gard. Herb., 12720, 6807); Utuado, N. L. Britton & J. F. Cowell, 999 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61492).

Jamaica: W. A. & E. L. Murrill, 40, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 56288); Castleton Gardens and Chester Vale, W. A. & E. L. Murrill, 52, 314, respectively, comm. by N. Y. Bot. Gard. Herb.; Hope Gardens, F. S. Earle, 178, comm. by N. Y. Bot. Gard. Herb.

Montserrat: Roches, J. A. Shafer, 915 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61473).

Argentina: R. Fries, 138, comm. by L. Romell, 333.

Philippine Islands: comm. by C. G. Lloyd, 11215 (in Mo. Bot. Gard. Herb., 58688).

Africa: Natal, Durban, P. A. van der Bijl, 2, 36 (in Mo. Bot. Gard. Herb., 58800, 58834); Unkomaas, P. A. van der Bijl, 1151 (in Mo. Bot. Gard. Herb., 62079).

10. C. bombycinum (Sommerf.) Bresadola, I. R. Accad. Agiati Atti III. 3: 111. 1897; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 233. 1911; Wakefield & Pearson, Brit. Myc. Soc. Trans. 6: 138. text f. 1919; Rea, Brit. Basid. 674. 1922.

Thelephora bombycina Sommerfelt, Fl. Lapp. Suppl. 284. 1826; Fries, Elench. Fung. 1: 211. 1828.

Type: in Sommerfelt Herb., in Univ. of Christiania Herb., a fragment in Burt Herb.

Fructifications irregularly effused, thick, membranaceous-soft, pieces separable, at first white, becoming pinkish buff to creambuff in the herbarium, even or varying rough to a hydnoid sur-

face, somewhat cracked, the margin and subiculum floccose to fibrillose and sometimes hirsute; in section 200–1000  $\mu$  thick, with the hyphae suberect, loosely interwoven, thick-walled, 4–5  $\mu$  in diameter, nodose-septate; no gloeocystidia; spores hyaline, even, 6–10  $\times$  5–6  $\mu$ .

Fructifications 3-10 cm. long, 2-3 cm. wide.

On bark of living and dead Salix and Alnus usually, but also on Betula, Acer, Tilia, Populus, and Pinus. In Europe and from Canada to Massachusetts and westward to Washington and Arizona, and in Texas. July to March. Uncommon.

C. bombycinum is a thick species with description somewhat suggestive of C. cremoricolor, but it does not crack radially, and tend to brown color like the latter, is more spongy and with more pelliculose hymenium and with a broader, thicker, and very conspicuous margin, and favors Salix as a substratum.

Specimens examined:

Exsiccati: Brinkmann, Westfälische Pilze, 11; Jaczewski, Fungi Rossiae, 232, under the name *Hypochnus Sambuci*; Romell, Fungi Scand., 35, under the name *Corticium serum*.

Norway: Saltd, Sommerfelt, fragment of type comm. by L. Romell.

Sweden: Stockholm, L. Romell, 63, 64, 65, 201, 344, and in Romell, Fungi Scand., 35; Upsala, L. Romell, two unnumbered specimens.

Russia: in Jaczewski, Fungi Rossiae, 232.

Germany: Lengerich, in Brinkmann, Westfälische Pilze, 11.

Austria: Feldkirch, Rick, comm. by Bresadola.

Canada: J. Macoun, 56, 60, in part, 157; Lower St. Lawrence Valley, J. Macoun, 30.

Ontario: Port Credit and Toronto, J. H. Faull, 655 and 380, respectively (in Mo. Bot. Gard. Herb., 44943, 44948).

Vermont: Middlebury, E. A. Burt.

Massachusetts: on beams in cotton mill, R. J. Blair, 248, in part, comm. by L. O. Overholts, 3812a (in Mo. Bot. Gard. Herb., 54995).

New York: Alcove, C. L. Shear, 1317; Clear Water, G. F. Atkinson, 5050; East Galway, E. A. Burt; Hudson Falls, S. H. Burnham, 14 (in Mo. Bot. Gard. Herb., 44007); Kenwood, S. H.

Burnham, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 56048).

Texas: Quitman, W. H. Long, 12092 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61343).

Michigan: Ann Arbor, C. H. Kauffman, 18.

Minnesota: Princeton, C. J. Humphrey, 1030 (in Mo. Bot. Gard. Herb., 21779).

Washington: Bingen, W. N. Suksdorf, 905, 915.

Arizona: Flagstaff, W. H. Long, 19449 (in Mo. Bot. Gard. Herb., 55141).

# 11. C. sociatum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, small, closely adnate, very thin, white, becoming continuous and somewhat waxy at the center, even, barely cracked, the margin thinning out, with hyphae interwoven; in section 70–90  $\mu$  thick, not colored, with the hyphae loosely interwoven near the substratum, 3  $\mu$  in diameter, not incrusted, not nodose-septate; no gloeocystidia; basidia with 4 sterigmata; spores hyaline, even,  $10\frac{1}{2}-12\times5-6~\mu$ , copious; a few imbedded spores present.

Fructifications 2-10 mm. long, 1-3 mm. wide—24 on an area 9 cm. long, 2 cm. wide.

On bark of decaying logs of *Thuja plicata*. Manitoba and British Columbia. August.

C. sociatum is a white species belonging in the group with C. arachnoideum, C. centrifugum, and C. pelliculare but distinct by the many small fructifications arranged near together, large spores, and hyphae neither nodose-septate nor incrusted.

Specimens examined:

Manitoba: Norway House, G. R. Bisby, 1466 (in Mo. Bot. Gard. Herb., 61649).

British Columbia: Kootenai Mts. near Salmo, J. R. Weir, 529, type (in Mo. Bot. Gard. Herb., 21596).

12. C. scutellare Berk. & Curtis, Grevillea 2: 4. 1873; Sacc. Syll. Fung. 6: 634. 1888; Massee, Linn. Soc. Bot. Jour. 27: 128. 1890.

Type: in Kew Herb. and Farlow Herb.

Fructifications long and widely effused, thin, adnate, from white becoming cream-buff to warm buff in the herbarium, waxy, often granular, finally very much cracked into minute areolae, 1–3 to a mm., which flake away from the substratum—sometimes leaving some of the white subiculum on the latter, the margin thinning out; in section 120–250  $\mu$  thick, not colored, composed of suberect, interwoven, thin-walled hyphae  $2\frac{1}{2}-3\frac{1}{2}\mu$  in diameter, incrusted in the subhymenial region so as to form a conspicuous subhymenial zone of mineral matter; no cystidia nor gloeocystidia; spores hyaline, even,  $4-6\times 2-3\mu$ .

Fructifications 2-8 cm. long, 1-4 cm. wide.

On fallen decaying limbs of frondose species. New York to Louisiana and westward to Kansas, in the West Indies, Japan, and South Africa. June to January. Common in the southern states.

C. scutellare, when fully mature in the southern states, may be recognized at sight by the very numerous areolae wholly separated from one another by fissures, but less mature and more northern specimens may be cracked into more rectangular masses up to 2 cm. in diameter and more or less connected together. In such specimens the subhymenial zone of mineral matter is a helpful character, for this zone is constant and conspicuous when sections are examined and, together with the small spores, afford sharp distinctive characters.

Specimens examined:

New York: Albany County, S. H. Burnham, 29 (in Mo. Bot. Gard. Herb., 54484); Alcove, C. L. Shear, 998; Fort Ann, S. H. Burnham, 43, in part (in Mo. Bot. Gard. Herb., 54453); Hudson Falls, S. H. Burnham, 16, 35 (in Mo. Bot. Gard. Herb., 54499, 54451); Ithaca, H. S. Jackson, C. Thom, comm. by Cornell Univ. Herb., 18201 and 14371, respectively; Karner, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb. 54380), and C. H. Peck, comm. by N. Y. State Mus. Herb., T6 (in Mo. Bot. Gard. Herb., 54640); Meadowdale, C. H. Peck, comm. by N. Y. State Mus. Herb., T6 (in Mo. Bot. Gard. Herb., 54640); North Elba, C. H. Peck (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 55973); Port Jefferson,

C. H. Peck (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 55981).

New Jersey: Newfield, J. B. Ellis, 418, 2052, 2475, and 2 unnumbered specimens comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 16061, 14255, 7657, 7456 and 44642, respectively).

Pennsylvania: Philadelphia, A. S. Rhoads, comm. by L. O. Overholts, 2680 (in Mo. Bot. Gard. Herb., 5918); Trexlertown, W. Herbst, 40.

District of Columbia: Takoma Park, E. M. Williams (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55812).

Virginia: Chain Bridge, A. S. Rhoads, comm. by L. O. Overholts, 3968 (in Mo. Bot. Gard. Herb., 54985).

South Carolina: type (in Curtis Herb., 2473).

Georgia: Tallulah Falls, A. B. Seymour, comm. by Farlow Herb., E (in Mo. Bot. Gard. Herb., 44610).

Florida: Mr. Curtiss, comm. by W. G. Farlow.

Alabama: Auburn, Ala. Biol. Survey; Montgomery, R. P. Burke, 6, 84, 143, 148, 239 (in Mo. Bot. Gard. Herb., 22316, 20508, 10673, 44907, 57104, respectively).

Mississippi: Ocean Springs, F. S. Earle, 183 (in Mo. Bot. Gard. Herb., 4838).

Louisiana: A. B. Langlois, 134, comm. by U. S. Dept. Agr. Herb.; St. Martinville, A. B. Langlois, aa, 856, 2632, and a specimen comm. by Lloyd Herb., 4128.

Kentucky: Mammoth Cave, C. G. Lloyd, 2562.

Indiana: Crawfordsville, D. Reddick, 9.

Illinois: Glencoe, E. T. & S. A. Harper, 821.

Missouri: Columbia, B. M. Duggar, 589.

Kansas: Rooks County, E. Bartholomew.

Jamaica: Chester Vale, W. A. & E. L. Murrill, 290, 329, 341, comm. by N. Y. Bot. Gard. Herb.; Hope Gardens, F. S. Earle, 192, comm. by N. Y. Bot. Gard. Herb.; Monkey Hill, W. A. & E. L. Murrill, 783, comm. by N. Y. Bot. Gard. Herb.; New Haven Gap, W. A. & E. L. Murrill, 766, comm. by N. Y. Bot. Gard. Herb.; St. Margaret's Bay, A. E. Wight, comm. by W. G. Farlow, 4 (in Mo. Bot. Gard. Herb., 44076).

Japan: Shinokubi, Prov. Harima, A. Yasuda, 6 (in Mo. Bot. Gard. Herb., 55664).

Africa: Erhove, Zululand, P. A. van der Bijl, 26 (in Mo. Bot. Gard. Herb., 58824); Houtbos, Transvaal, P. A. van der Bijl, 1482.

13. C. tuberculatum Karsten, Hedwigia 35: 45. 1896; Krit. Öfvers. Finl. Basidsv. Tilläg 3: 29. 1898; Sacc. Syll. Fung. 14: 221. 1899; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1561. 1906.

Type: authentic specimen or part of type in Burt Herb.

Fructifications orbicular or longitudinally effused, rather thick, somewhat membranaceous, small pieces separable when moistened, white at first, becoming light buff to warm buff in the herbarium, somewhat colliculose or tuberculate, waxy, the margin radiately fibrillose; in section 200–300  $\mu$  thick, not colored, with the hyphae densely interwoven in a narrow layer next to the substratum and then ascending obliquely and not crowded to the compact hymenial layer,  $3\frac{1}{2}-4\frac{1}{2}\mu$  in diameter, somewhat incrusted in the type, not nodose-septate; no gloeocystidia; spores hyaline, even,  $4-6 \times 2\frac{1}{2}-3\frac{1}{2}\mu$ , copious.

Pieces of fructification  $2\frac{1}{2}$  cm. in diameter in the specimen seen.

On bark and wood of fallen branches of *Populus*, *Fraxinus*, and other frondose species. Finland, Pennsylvania to Wisconsin. Rare.

C. tuberculatum approaches Radulum in having a middle layer of loosely arranged, ascending hyphae and a somewhat colliculose surface and some small tubercules in the authentic specimen communicated to me by Karsten and which agrees closely with his description of the species. The general aspect somewhat resembles that of Peniophora mutata. The American gatherings cited below have a more even hymenium and hyphae not incrusted and are doubtfully referred to C. tuberculatum.

Specimens examined:

Finland: Mustiala, P. A. Karsten, probably part of type.

Pennsylvania: Trexlertown, W. Herbst, 77.

Michigan: East Tower, J. R. Weir, 370 (in Mo. Bot. Gard. Herb., 17074).

Wisconsin: Madison, A. O. Stucki, 44, comm. by Univ. Wis. Herb.

14. C. crustaceum (Karsten) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1566. 1906.

Xerocarpus crustaceus Karsten, Hedwigia 35: 45. 1896.— Stereum crustaceum Karsten in Sacc. Syll. Fung. 14: 215. 1899. Type: in Karsten Herb. and Burt Herb.

Fructifications effused, thin, crustaceous-adnate, somewhat grumose, not at all separable, white or whitish, even or somewhat granular, conforming to inequalities of the substratum, somewhat cracked; in section 40–100  $\mu$  thick, not colored, composed of densely arranged hyphae 2  $\mu$  in diameter, not well shown, with crystalline masses intermixed; no gloeocystidia; spores hyaline, even,  $4\frac{1}{2}-5 \times 3 \mu$ , copious.

Fructifications 2-6 cm. long, 1-3 cm. wide.

On rough bark of Acer, Crataegus, Populus, Salix, Ulmus, and Abies. Finland and Canada to Florida. July to November. Probably common.

C. crustaceum is so similar in aspect to Peniophora Sambuci that it is necessary to distinguish it from the latter by the microscopic characters of sectional preparations. C. crustaceum has no cystidia, has more densely arranged hyphae and a good deal of obscuring crystalline matter intermixed.

Specimens examined:

Finland: Mustiala, P. A. Karsten, authentic specimen on Populus. Canada: J. Macoun, 1, 2; St. Lawrence Valley, J. Macoun, 27, 49, 51.

Ontario: Ottawa, J. Macoun (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55902), and 4.

Quebec: Hull, J. Macoun, 82.

Vermont: Middlebury, E. A. Burt.

West Virginia: Paw Paw, C. L. Shear, 1176.

Florida: Jacksonville, W. W. Calkins, comm. by Farlow Herb. (in Mo. Bot. Gard. Herb., 44637).

15. C. pelliculare Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 48: 411. 1889; Hedwigia 35: 46. 1896; Sacc. Syll. Fung. 9: 232. 1891; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 239. 1911.—Cf. v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1556. 1906.

Type: fragment of type and authentic specimen in Burt Herb. Fructifications broadly effused, thin, membranaceous, tender, small pieces separable, white when fresh, becoming ivory-yellow to cream-buff in the herbarium, even, somewhat cracked and showing a loose, cottony subiculum which extends out beyond the hymenium as a fimbriate, white margin; in section  $100-300 \,\mu$  thick, not colored, composed of loosely interwoven and ascending, thin-walled hyphae  $2\frac{1}{2}-3\frac{1}{2}\mu$  in diameter, sparingly nodose-septate, rarely incrusted in the subhymenium; no gloeocystidia; spores hyaline, even,  $4-6 \times 2-3 \,\mu$ .

Fructifications 2-6 cm. long, 1-3 cm. wide.

On decaying limbs of both coniferous and frondose species. In Europe and from New Hampshire to Pennsylvania, in Illinois, British Columbia to Mexico, and in Bermuda. June to December. Infrequent.

P. pelliculare has delicate white to creamy fructifications distinguishable from those of C. lacteum by the small spores not at all globose.

Specimens examined:

Exsiccati: Thümen, Myc. Univ., 1607, under the name Corticium laeve.

Finland: Mustiala, P. A. Karsten, fragment of type comm. by Karsten to Bresadola and by Bresadola to Romell and by Romell to Burt.

Sweden: K. Starback, authentic specimen comm. by Karsten; L. Romell, 319; Femsjö, E. A. Burt, two gatherings; Stockholm, L. Romell, 298A, 320.

New Hampshire: Chocorua, W. G. Farlow, C37 (in Mo. Bot. Gard. Herb., 43968).

Vermont: Middlebury, E. A. Burt.

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and in Mo. Bot. Gard. Herb., 57490); Orient Point, R. Latham, 3 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55700).

New Jersey: Belleplain, C. L. Shear, 1237; Newfield, J. B. Ellis, in Ell. & Ev. Fungi Col., 1207.

Pennsylvania: Bear Meadows, L. O. Overholts, 2890 (in Mo. Bot. Gard. Herb., 5717); Trexlertown, W. Herbst, 15.

Michigan: Ann Arbor, C. H. Kauffman, 19.

Illinois: Helleydayboro, C. J. Humphrey, 1351 (in Mo. Bot. Gard. Herb., 59017); Port Byron, E. T. & S. A. Harper, 733.

British Columbia: Kootenai Mts., Salmo, J. R. Weir, 456 (in Mo. Bot. Gard. Herb., 13043); Sidney, J. Macoun, 11 (in Mo. Bot. Gard. Herb., 5729).

Washington: Bingen, W. N. Suksdorf, 879, 919.

Arizona: Flagstaff, W. H. Long, 19491 (in Mo. Bot. Gard. Herb., 44738, 55135); First Valley Experiment Station, W. H. Long, 21119 (in Mo. Bot. Gard. Herb., 55136).

Mexico: Chihuahua, Parral, E. O. Mathews, 2, 26 (in Mo. Bot. Gard. Herb., 44126, 44125); Guernavaca, W. A. & E. L. Murrill, 418, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb. 54512).

Bermuda: on cornstalks, S. Brown, N. L. Britton & F. J. Seaver, 11248 (in N. Y. Bot. Gard. Herb. and Mo. Bot. Gard. Herb., 4809).

C. Auberianum Montagne in La Sagra, Hist. de Cuba 9<sup>2</sup>:
 1845; Syll. Crypt. 178. 1856; Sacc. Syll. Fung. 6: 616.
 1888; Massee, Linn. Soc. Bot. Jour. 27: 135. 1890.

Type: part of type in Kew Herb.

Fructifications effused, orbicular at first, becoming longitudinally elongated, adnate, very thin, white, floccose-farinaceous, even, sometimes somewhat cracked, the margin thinning out, floccose; in section 45–120  $\mu$  thick, not colored, composed of suberect, branching, interwoven, thin-walled hyphae about 2  $\mu$  in diameter, not nodose-septate; no gloeocystidia; no cystidia; spores hyaline, even, flattened on one side, 4–5  $\times$  2–3  $\mu$ .

Fructifications at first 2-10 mm. in diameter, finally up to 10 cm. long, 1 cm. broad.

On small decaying, fallen twigs of frondose species. Vermont to Louisiana, and in the West Indies. August to March. Rare.

C. Auberianum may be recognized by its very thin, snow-white fructifications having a farinose hymenial surface, small spores, and slender, thin-walled hyphae throughout. No gloeocystidia are present nor coarse hyphae near substratum. The occurrence of several small fructifications near together when young is characteristic. The hyphae are probably somewhat incrusted, but this needs confirmation.

Specimens examined:

Vermont: E. A. Burt, two gatherings.

North Carolina: Blowing Rock, G. F. Atkinson, 4330.

South Carolina: On Carya, Curtis Herb., 2497 (in Kew Herb.).

Georgia: Tallulah Falls, A. B. Seymour, comm. by Farlow Herb., DD (in Mo. Bot. Gard. Herb., 44595).

Florida: Sands Key, R. A. Harper, 6 (in Mo. Bot. Gard. Herb., 54537).

Louisiana: St. Martinville, A. B. Langlois, Q, R.

Arkansas: Womble, W. H. Long, 19823, 19821, in part (in Mo. Bot. Gard. Herb., 8633, 17801).

Bermuda: Walsingham, H. H. Whetzel, Aat (in Mo. Bot. Gard. Herb., 58718).

Cuba: presumable part of type from Montagne to Berkeley (in Kew Herb.); Managua, Earle & Murrill, 26, comm. by N. Y. Bot. Gard. Herb.; San Antonio de los Baños, Havana Province, Earle & Murrill, 46, comm. by N. Y. Bot. Gard. Herb.; San Diego de los Baños, Havana Province, Earle & Murrill, 332, comm. by N. Y. Bot. Gard. Herb.; locality not stated, C. G. Lloyd, 430 (in Mo. Bot. Gard. Herb., 55176).

17. C. galactinum (Fr.) Burt, in Moffatt, Chicago Acad. Sci. Bul. 7: 137. 1909.

Thelephora galactina Fries, R. Soc. Sci. Upsal. Acta III. 1:136. 1851; Sacc. Syll. Fung. 6: 541. 1888; von Schrenk, Bot. Gaz. 34:65. 1902.—An Corticium rigescens Berk. & Curtis in Cooke, Grevillea 20: 12. 1891?

Type: in Fries Herb. and Curtis Herb.

Fructifications long and broadly effused, becoming rather thick, coriaceous-soft, closely adnate, small pieces separable, white to cream-color, waxy, even, not cracked, the margin indeterminate, thinning out, with the hyphae interwoven; in section 200–1000  $\mu$  thick, not colored, composed of suberect, densely interwoven, hyaline hyphae about 1–2  $\mu$  in diameter, not incrusted; no gloeocystidia; curved ends of the hyphae or their branches form the surface of the hymenium and are about  $\frac{1}{2}$ –1  $\mu$  in diameter; spores white in spore collection,  $4-5\frac{1}{2}\times 2-3$   $\mu$ .

Fructifications 4-12 cm. long, 2-4 cm. wide.

On roots of living apple and blackberry plants, on the ground, and broadly effused on rotting logs of frondose and coniferous species. Canada to Texas and westward to the Pacific coast, in West Indies and in Japan. Throughout the year. Common.

C. galactinum resembles C. portentosum in aspect but has a more erect hyphal structure and is usually not at all stratose and with substance not colored. Both species have a hymenial surface composed of fine, curved hyphal branches, but the spores of C. galactinum are smaller and ellipsoid and those of C. portentosum spherical. The mycelium of C. galactinum was collected as a parasitic root rot on the roots of young apple trees and blackberry bushes and developed mature fructifications. The collector's data on the type specimen of this species is "In radicibus ad latera fossarum."

Specimens examined:

- Exsiccati: Ravenel, Fungi Car. 4: 15, under the name Corticium calceum.
- Canada: J. Macoun, 26, 31, 111; Lower St. Lawrence Valley, J. Macoun, 4, 9, 35, 83.
- Ontario: Lake Rosseau, E. T. & S. A. Harper, 638, 640; Nixon, J. Dearness, 1023 (in Mo. Bot. Gard. Herb., 22732); Ottawa, J. Macoun, 56, 248, in part; Temagami, H. von Schrenk (in Mo. Bot. Gard. Herb., 57053).
- Maine: New Limerick, H. von Schrenk, 62 (in U. S. Dept. Agr. Herb. and Burt Herb.); Piscataquis County, W. A. Murrill, 1881 (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 61423, and Burt Herb.).
- New Hampshire: Chocorua, W. G. Farlow (in Mo. Bot. Gard. Herb., 19544); North Conway, L. O. Overholts, 4555, 4584 (in Mo. Bot. Gard. Herb., 55635, 55634).
- Vermont: Grand View Mt., E. A. Burt; Little Notch, E. A. Burt; Middlebury, E. A. Burt, five gatherings.
- New York: Arkville, W. A. Murrill (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61362, 61363); Cranberry Lake, A. H. W. Povah, 772 (in Mo. Bot. Gard. Herb., 3730); East Galway, E. A. Burt; Floodwood, E. A. Burt, C. H. Peck, 12; Forestburgh, C. H. Peck, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 56049); Freeville, G. F. Atkinson,

18186; Gansevoort, C. H. Peck, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 55982); Ithaca, G. F. Atkinson, 2869, 4898; Jenkinsville, S. H. Burnham, 40 (in Mo. Bot. Gard. Herb., 54452); Karner, H. D. House, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 55197); Lake Placid, W. A. & E. L. Murrill, 270 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61578); North Elba, C. H. Peck, 12, and (in N. Y. State Mus. Herb., T 26, and Mo. Bot. Gard. Herb., 54652); Oneida, H. D. House, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 57414); Pompey, L. M. Underwood, 25, 107, 357 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61432, 61431, 61575); West Fort Ann, S. H. Burnham, 13, in part (in Mo. Bot. Gard. Herb., 54505); White Plains, L. M. Underwood (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61574).

Pennsylvania: Mt. Gretna, E. A. Burt; State College, L. O. Overholts, 4711 (in Mo. Bot. Gard. Herb., 56116).

Virginia: Woodstock, C. L. Shear, 1195.

South Carolina: H. W. Ravenel, in Ravenel, Fungi Car. 4: 15, on pine, and type (in Fries Herb., and Curtis Herb., 1601).

Florida: W. W. Calkins (in U. S. Dept. Agr. Herb., and Burt Herb.); Starke, C. L. Shear, 2904 (in Mo. Bot. Gard. Herb., 15311); Tallahassee, E. Bartholomew, 5708 (in Mo. Bot. Gard. Herb., 44255).

Louisiana: Bogalusa, C. J. Humphrey, 5516; St. Martinville, A. B. Langlois, 607 (in U. S. Dept. Agr. Herb., and Burt Herb.), 1762, X.

Texas: Houston, H. W. Ravenel, 268 (in U. S. Dept. Agr. Herb., and Burt Herb.).

West Virginia: Eglon, C. G. Lloyd, 02643.

Ohio: Cincinnati, A. P. Morgan, comm. by Lloyd Herb., 2604.

Illinois: on apple roots, H. von Schrenk; River Forest, E. T. & S. A. Harper, 655.

Michigan: Mass, C. J. Humphrey, 1583 (in Mo. Bot. Gard. Herb., 10743); Three Lakes, C. J. Humphrey, 1602 (in Mo. Bot. Gard. Herb., 17883); Vermilion, A. H. W. Povah, 203 (in Mo. Bot. Gard. Herb., 15326).

Missouri: Grandin, H. von Schrenk (in Mo. Bot. Gard. Herb.,

- 43022); St. Louis, on apple roots, *H. von Schrenk*, three gatherings.
- Arkansas: on blackberry roots, G. M. Darrow (in Mo. Bot. Gard. Herb., 63734); on apple roots, H. von Schrenk; Fordyce, C. J. Humphrey, 5812; Womble, W. H. Long, 19816, 19838, 19864, 21104 (in Mo. Bot. Gard. Herb., 8958, 8634, 8635, 55144).
- Colorado: Golden, L. O. Overholts, 1745 (in Mo. Bot. Gard. Herb., 54874).
- Montana: Como, E. E. Hubert, comm. by J. R. Weir, 11959 (in Mo. Bot. Gard. Herb., 63316); Evaro, J. R. Weir, 437 (in Mo. Bot. Gard. Herb., 14387); Rexford, E. E. Hubert, comm. by J. R. Weir, 11977 (in Mo. Bot. Gard. Herb., 63330).
- Idaho: Coeur d'Alene, E. E. Hubert, comm. by J. R. Weir, 12002 (in Mo. Bot. Gard. Herb., 63364); Coolin, J. R. Weir, 11504 (in Mo. Bot. Gard. Herb., 63285); Priest River, J. R. Weir, 15, and 133, 346 (in Mo. Bot. Gard. Herb., 12119, 7561), and E. E. Hubert, comm. by J. R. Weir, 12025 (in Mo. Bot. Gard. Herb., 63378); St. Maries, J. R. Weir, comm. by C. J. Humphrey, 2556 (in Mo. Bot. Gard. Herb., 13030), and E. E. Hubert, comm. by J. R. Weir, 11997 (in Mo. Bot. Gard. Herb., 63360).
- Manitoba: Winnipeg, G. R. Bisby & I. L. Conner, 1102 (in Mo. Bot. Gard. Herb., 59038).
- British Columbia: Kootenai Mts., near Salmo, J. R. Weir, 457, 500, 508, 531, 542 (in Mo. Bot. Gard. Herb., 9122, 21631, 20270, 23118, 14254).
- Washington: Chehalis, C. J. Humphrey, 6289 (in Mo. Bot. Gard. Herb., 10751); Lake Wilderness, C. H. Kauffman, 17 (in Mo. Bot. Gard. Herb., 4674); Renton, C. J. Humphrey, 6640; Sedro Woolley, C. J. Humphrey, 7568 (in Mo. Bot. Gard. Herb., 10775).
- Cuba: Ceballos, C. J. Humphrey, 2730 (in Mo. Bot. Gard. Herb., 9083).
- Porto Rico: Rio Piedras, J. A. Stevenson, 1195, 3224 (in Mo. Bot. Gard. Herb., 6949, 7734).
- Japan: Hiroto-Mura, Prov. Awaji, A. Yasuda, 24 (in Mo. Bot. Gard. Herb., 55662); Mt. Mikuma, Prov. Awaji, A. Yasuda, 17 (in Mo. Bot. Gard. Herb., 55661).

#### 18. C. calceum Fries emend. Romell & Burt

C. calceum Fries, Epicr. 562. 1838, in part; Hym. Eur. 652. 1874, in part; Sacc. Syll. Fung. 6: 622. 1888, in part.—The-lephora calcea Fries var. glebulosa Fries, Elench. Fung. 2: 215. 1828.—Not Peniophora glebulosa Bresadola, Fungi Trid. 2: 61 pl. 170, f. 2. 1898.

Type: in Fries Herb. and a fragment in Burt Herb.

Fructifications broadly effused, very thin, closely adnate, not at all separable, floccose-membranaceous, white, sometimes becoming ivory-yellow in the herbarium, even, cracking to the substratum into small rectangular masses 1-4 to a mm., the margin farinose; in section 100–200  $\mu$  thick, not colored, with the hyphae erect, densely crowded together and interwoven, somewhat conglutinate, short-celled,  $1-1\frac{1}{2}$   $\mu$  in diameter, sometimes with algal cells imbedded; no gloeocystidia nor cystidia; spores hyaline, even,  $3-5\frac{1}{2} \times 1\frac{1}{2}-2$   $\mu$ .

Fructifications 3-20 cm. long, 1-5 cm. wide.

Under side of decaying rails of *Pinus sylvestris* and *P. Strobus*, and on decaying wood of logs of *P. monticola* and *Thuja*. In Sweden and from Vermont and New Jersey to Idaho and British Columbia. July to November. Abundant when found.

Since C. calceum var. glebulosum is all that now remains under C. calceum after the segregation under other names of all other components, no confusion should result from the present proposed restriction of the species C. calceum. It may be added that the original description of C. calceum applies better to the emended species than to any of the other components withdrawn. dola studied the Friesian type of Thelephora calcea var. glebulosa and identified it with Peniophora glebulosa, a species very common throughout Europe. He shared a portion of his Friesian type with me and accompanied it with notes on microscopic details in which he stated, "Cystidia adsunt sed collapsa." However, no cystidia are present in this fragment, nor in the type in Fries Herb., nor in ample collections of the species made by Romell and myself at the type station, Femsjö. I have not been able to recognize this species in the extensive series of Corticiums received from countries of Europe other than Sweden. Since the species is widely distributed and abundant in northern United States, it is possible that it is a North American endemic species which became established in Sweden as an outlying station, comparable with cases of *Stereum rufum*, *Stereum Murrayi*, etc.

Specimens examined:

Exsiccati: Ell. & Ev., N. Am. Fungi, 2807, under the name Corticium scutellare; Ell. & Ev., Fungi Col., 104, under the name Corticium scutellare.

Sweden: Femsjö, E. Fries, type of Thelephora calcea var. glebulosa (in Fries Herb., and fragment in Burt Herb.), L. Romell, 185, 211, and Romell & Burt, two gatherings; Stockholm, L. Romell, 321, 322, 324, 325.

Canada: J. Macoun, 1, 34; Lower St. Lawrence Valley, J. Macoun, 57.

Vermont: Middlebury, E. A. Burt, two gatherings.

New York: Bolton, C. H. Peck, 9; Clearwater, G. F. Atkinson, 5046; Floodwood, C. H. Peck, 11; Ithaca, G. F. Atkinson, 941, 22972; Schuylerville, C. H. Peck, 20.

New Jersey: Newfield, J. B. Ellis, in Ell. & Ev., N. Am. Fungi, 2807, and in Ell. & Ev., Fungi Col., 104.

Pennsylvania: State College, L. O. Overholts, 4809 (in Mo. Bot. Gard. Herb., 56119).

Michigan: Mass, C. J. Humphrey, 1662 (in Mo. Bot. Gard. Herb., 17607).

Wisconsin: Lake Glencoe, E. T. & S. A. Harper, 853.

Idaho: Priest River, J. R. Weir, 40, 64, and 6350 (in Mo. Bot. Gard. Herb., 58387).

British Columbia: Kootenai Mts., near Salmo, J. R. Weir, 461, 463, 533 (in Mo. Bot. Gard. Herb., 9119, 12631, 20973).

# 19. C. vescum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, not at all separable, from white to pale drab-gray in the herbarium, even, not shining, not cracked, the margin thinning out, indeterminate; in section 20-30  $\mu$  thick, not colored, very compact, composed of very short, erect hyphae which terminate in basidia; no gloeocystidia; spores hyaline, even, allantoid,  $4\frac{1}{2} \times \frac{1}{2}-1$   $\mu$ .

Fructifications up to 6 cm. long, 3 cm. wide.

On decorticated pine limb completely decayed by a brittle, brown rot. Maryland and Alabama. October.

C. vescum looks like a thin, whitish or somewhat cinereous wash in water color on the surface of the weathered pine limb. No interwoven hyphal structure is visible under a lens, for the short basal hyphae start out vertically from the substratum and terminate in basidia packed closely together in the hymenium.

Specimens examined:

Maryland: Takoma Park, C. L. Shear, 961.

Alabama: Montgomery, R. P. Burke, 476, type (in Mo. Bot. Gard. Herb., 57294).

#### 20. C. incanum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, very thin, closely adnate, not separable, becoming pearl-gray to mineral-gray in the herbarium, even, waxy, not cracked, the margin thinning out, indeterminate; in section 20–75  $\mu$  thick, not colored, composed of densely interwoven, hyaline hyphae 2½–3  $\mu$  in diameter, rarely nodose-septate, not incrusted; no gloeocystidia; basidia simple, with 4 short, blunt sterigmata; spores hyaline, even, about  $3½–4 \times 1½–3~\mu$ .

Fructifications 4-8 cm. long, 1-2 cm. wide.

On bark and wood of dead *Acer* and other frondose limbs. Canada to North Carolina. October and November.

C. incanum forms a thin, inseparable coating of mineral-gray color over bark and wood of frondose species usually. The aspect is so similar to that of common *Peniophora cinerea* that it is likely to be passed by as the latter, if examination of microscopic structure is not made.

Specimens examined:

Canada: J. Macoun, 36; Ottawa, J. Macoun, 32, 34.

Vermont: Middlebury, E. A. Burt.

New York: Karner, H. D. House, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 54384).

New Jersey: Belleplain, C. L. Shear, 1249, type.

North Carolina: Chapel Hill, J. N. Couch, 4225, comm. by W. C. Coker, under the name C. ochraceo-niveum (in Mo. Bot. Gard. Herb., 57412).

#### 21. C. canum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, thin, hypochnoid, tender, not separable, whitish to pale pinkish buff in the herbarium, even, with the hymenium fibrillose under a lens rather than in the form of a continuous pellicle, the margin thinning out, arachnoid; in section 100–180  $\mu$  thick, not colored, composed of lax, loosely interwoven hyphae  $2\frac{1}{2}\mu$  in diameter, thin-walled, nodose-septate, not incrusted, bearing a more compact hymenium; no gloeocystidia; spores hyaline, even,  $3-4 \times 1\frac{1}{2}-2\mu$ .

Fructifications 3-5 cm. long, \(\frac{1}{2}-1\frac{1}{2}\) cm. wide.

On decaying wood and bark of conifers. Canada to Louisiana and in Washington. September to October. Infrequent.

C. canum belongs in the group with C. centrifugum and C. pelliculare but differs from both in more hypochnoid structure and smaller spores. The hyphae are nodose-septate and not incrusted.

Specimens examined:

Canada: J. Macoun, 13, type, and 86, in part.

New York: Ithaca, G. F. Atkinson, 2563.

Maryland: Takoma Park, C. L. Shear, 1063.

Louisiana: St. Martinville, A. B. Langlois, 168, comm. by Lloyd Herb., 3046.

Idaho: Coolin, J. R. Weir, 11101 (in Mo. Bot. Gard. Herb., 63391); Priest River, J. R. Weir, 21.

British Columbia: Salmo, Kootenai Mts., J. R. Weir, 447 (in Mo. Bot. Gard. Herb., 21800).

Washington: Hoquiam, C. J. Humphrey, 6375, 6413.

C. centrifugum (Lév.) Bresadola, Ann. Myc. 1:96. 1903;
 Höhnel, Ann. Myc. 3: 188. 1905; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 240. 1911.

Rhizoctonia centrifuga Léveillé, Ann. Sci. Nat. Bot. II. 20: 225. 1843.—Hypochnus centrifugus Tulasne, Fung. Carp. 1: 113. 1861; Sacc. Syll. Fung. 6: 654. 1888.—Corticium decipiens v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 117: 1116. 1908.

Fructifications effused, very thin, arachnoid, forming a con-

tinuous hymenial pellicle, fragile, white, becoming pale olivebuff in the herbarium, even, the margin arachnoid or byssoid; in section 75–150  $\mu$  thick, not colored, with the hyphae loosely interwoven, thin-walled, not incrusted, usually 2–3  $\mu$  in diameter, sometimes with a few coarser and up to 6  $\mu$  in diameter along the substratum, only rarely nodose-septate; no gloeocystidia; spores hyaline, even, ellipsoidal, 4–8  $\times$  2½–4  $\mu$ .

Fructifications 2-6 cm. long, 1-3 cm. wide.

On decaying wood and leaves and fallen branches. Common in Europe, infrequent from Canada to Louisiana and westward to the Pacific and in the West Indies. June to February.

C. centrifugum is related to C. arachnoideum and C. pelliculare. Its more elongated spores, thinner and less arachnoid fructifications, and hyphae with only very few clamp connections separate it from C. arachnoideum, while C. pelliculare becomes more yellow in the herbarium, is likely to show some hyphal incrustation, and has rather smaller spores and a more compact hymenium. According to the original description C. decipiens differs by not having clamp connections but they are certainly present in the authentic specimen communicated by Litschauer.

Specimens examined:

Exsiccati: Ell. & Ev., Fungi Col., 309, under the name Corticium arachnoideum.

Sweden: L. Romell, 76; Stockholm, L. Romell, 60, 61, 168, 296, 348.

Germany: W. Brinkmann, comm. by Bresadola.

Austria: Klosterberg, Tirol, V. Litschauer, and another specimen under the name C. decipiens, determined and comm. by Litschauer.

Canada: Ottawa, J. Macoun, 49, 52.

Maine: Kittery Point, R. Thaxter (in Mo. Bot. Gard. Herb., 57606).

New Hampshire: Chocorua, W. G. Farlow; Shelburne, W. G. Farlow, 2.

New York: East Galway, E. A. Burt; Ithaca Flats, C. O. Smith, comm. by G. F. Atkinson, 8226; Karner, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 54350).

New Jersey: Newfield, J. B. Ellis, in Ell. & Ev., Fungi Col., 309.

Pennsylvania: State College, L. O. Overholts, 3630 (in Mo. Bot. Gard. Herb., 54698).

District of Columbia: Takoma Park, C. L. Shear, 1347.

Louisiana: St. Martinville, A. B. Langlois, ay.

Manitoba: Stony Mountain, A. H. R. Buller, 900 (in Mo. Bot. Gard. Herb., 58999); Winnipeg, G. R. Bisby, 1342 (in Mo. Bot. Gard. Herb., 60551).

Washington: Bingen, W. N. Suksdorf, 914.

Oregon: Corvallis, S. M. Zeller, 2066 (in Mo. Bot. Gard. Herb., 58767).

California: Massack, A. S. Rhoads, 18 (in Mo. Bot. Gard. Herb., 56987).

Jamaica: Castleton Gardens, W. A. & E. L. Murrill, 67, comm. by N. Y. Bot. Gard. Herb.; Chester Vale, W. A. & E. L. Murrill, 372, comm. by N. Y. Bot. Gard. Herb.

#### 23. C. Atkinsonii Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, adnate, thin, small pieces separable when moistened, white, even, waxy, not cracked, the margin thinning out, with hyphae interwoven; in section about 150  $\mu$  thick, not colored, composed of interwoven, branching, thin-walled, occasionally nodose-septate hyphae 3  $\mu$  in diameter, not incrusted, which have in the middle and subhymenial region an additional branched system of branches not more than 1  $\mu$  in diameter and bearing short acicular branchlets; no gloeocystidia; basidia simple, usually 4 sterigmata but rarely 5 or 6; spores hyaline, even,  $4\frac{1}{2} \times 2-2\frac{1}{2} \mu$ .

Fructifications 1-3 cm. long, 1-2 cm. wide.

On decaying, charred frondose wood and on *Populus*. New York and Louisiana. November and January.

C. Atkinsonii has snow-white color, waxy surface and small spores. The noteworthy character separating it from other white species is the system of delicate hyphal branches, so abundant in the middle and subhymenial regions of the fructification that they mask the outlines of the usual hyphae there and so fine that on first impression they seem to be the walls of collapsed hyphae. The mode of branching is not exactly that of C. in-

vestiens and C. jamaicense but a comparable type of hyphal differentiation. The great distance between the two stations leads me to suspect that C. Atkinsonii is more frequent than indicated by the collections in which the distinctive branching was observed.

Specimens examined:

New York: Altamont, E. A. Burt; Ithaca, G. F. Atkinson, 2558, type.

Louisiana: A. B. Langlois, 246, comm. by U. S. Dept. Agr. Herb.

#### 24. C. subnullum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, cartridge-buff to olive-buff in the herbarium, hypochnoid, not forming a continuous hymenium but with the basidia in more or less connected tufts of about 3–5 to the mm., farinaceous, the margin similar; in section 30–45  $\mu$  thick, not colored, composed of loosely arranged, hyaline hyphae 2–2½  $\mu$  in diameter, thin-walled, not incrusted, not nodose-septate; no gloeocystidia; spores hyaline, even, globose,  $2\frac{1}{2}$   $\mu$  in diameter, borne 4 to a basidium.

Fructifications 3-7 cm. long, 2-3 cm. wide.

On bark of decaying logs of *Populus* sp. British Columbia. July.

When C. subnullum becomes better known from additional collections, it may become necessary to transfer it to another genus, but the present gathering favors the view that it is a Corticium somewhat lacking basidia so that the hymenium becomes discontinuous. This character, occurrence on poplar bark, small spores, and general aspect of an olive-buff Hyphomycete are good distinctive characters.

Specimens examined:

British Columbia: Sidney, J. Macoun, 30, type (in Mo. Bot. Gard. Herb., 63776).

# 25. C. crustulinum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, thin, tender, separable, with the substance whitish, dry, soft and cottony, and the hymenium warm

buff in the herbarium, even, pelliculose, brittle, not shining, the margin whitish, continuous with the substance, fimbriate; in section 160  $\mu$  thick, not colored, composed of a layer next the substratum of loosely interwoven, hyaline, thin-walled hyphae 2  $\mu$  in diameter, nodose-septate, not incrusted, and of a compactly interwoven, thin hymenium; no gloeocystidia; basidia 6  $\times$  3  $\mu$ , with 4 sterigmata; spores hyaline, even, subglobose, 3  $\times$  2–3  $\mu$ , copious.

Fructifications 5 cm. long, 2-3 cm. wide.

On very rotten frondose wood. Porto Rico. July.

C. crustulinum is characterized by the loosely attached, whitish-margined fructifications with yellowish hymenium borne on a white, cottony substance. The small hyphae, small basidia, and small spores are good confirmatory characters. We have no closely related species.

Specimens examined:

Porto Rico: Rio Piedras, J. A. Stevenson, 2914, type (in Mo. Bot. Gard. Herb., 3130).

26. C. tessulatum Cooke, Grevillea 6: 132. 1878; Sacc. Syll.
Fung. 6: 619. 1888; Massee, Linn. Soc. Bot. Jour. 27: 136. 1890.
Type: type distribution in Ravenel, Fungi Am., 127.

Fructifications effused, adnate, thin, somewhat membranaceous, tender, small pieces separable, in the herbarium becoming naphthalene-yellow, with central parts light ochraceous buff, even, contracting greatly in drying, and cracking into rectangular masses 1–4 mm. in diameter separated by fissures 1–2 mm. wide, with some of the white silky subiculum clinging to the substratum, the margin whitish, fibrillose; in section 150–200  $\mu$  thick, not colored, composed of loosely interwoven, very thin-walled and collapsing hyphae 4  $\mu$  in diameter, abundantly nodose-septate, not incrusted; no gloeocystidia; spores hyaline, even,  $4-4\frac{1}{2}\times3$   $\mu$ , few found.

Fructifications 2-4 cm. in diameter.

On pine and spruce bark on the ground. Canada to South Carolina, and in Idaho and Arizona. May to October. Infrequent.

C. tessulatum is somewhat suggestive of C. Berkeleyi in aspect

but is colored differently, tending towards light ochraceous-buff in the more central parts of the fructification; this color, occurrence on old pine and spruce, the wide cracks from drying, and loose attachment to substratum and tendency to scale away from it of the rectangular masses of the dried fructification are helpful characters in recognizing the species. *C. illaqueatum*, occurring on *Castanea* in France, is closely related.

Specimens examined:

Exsiccati: Ravenel, Fungi Am., 127, type distribution.

Canada: Lower St. Lawrence Valley, J. Macoun, 71, 75; Ontario, Temagami, H. von Schrenk (in Mo. Bot. Gard. Herb., 57051).

Maine: Penobscot County, W. A. Murrill, 1821 (in N. Y. Bot. Gard. Herb., Burt Herb., and Mo. Bot. Gard. Herb., 59676).

New Hampshire: Chocorua, W. G. Farlow, 10, and two other gatherings.

Vermont: Middlebury, E. A. Burt.

New York: Osceola, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 59674, 59676).

Maryland: Takoma Park, C. L. Shear, 1066.

South Carolina: Aiken, H. W. Ravenel, in Ravenel, Fungi Am., 127.

Idaho: Addie, E. E. Hubert, comm. by J. R. Weir, 11989 (in Mo. Bot. Gard. Herb., 63352).

Arizona: Flagstaff, W. H. Long, 19494 (in Mo. Bot. Gard. Herb., 44768, 44769); Interior Basin, San Francisco Peaks, W. H. Long, 21309, in part (in Mo. Bot. Gard. Herb., 54890).

# 27. C. Stevensonii Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, rather thick, fleshy-membranaceous, small pieces separable, becoming cartridge-buff to cream-buff in the herbarium, perhaps white when growing, ceraceous, slightly colliculose, becoming somewhat cracked in drying, the margin narrow, similar; in section 400–450  $\mu$  thick, not colored, with an incrusted subhymenial zone, the hyphae 3–3½  $\mu$  in diameter, not nodose-septate, rather thick-walled and rigid, loosely interwoven and rising obliquely to the base of the compact subhymenium, conspicuously incrusted for a length of about 30  $\mu$  in the incrusted

zone and about 6  $\mu$  in diameter over the incrustation; no gloeocystidia; spores copious, hyaline, even,  $6 \times 4-4\frac{1}{2}\mu$ .

Fructifications in fragments 1-3 cm. long, 1-2 cm. wide.

On badly decayed frondose wood. Porto Rico. December.

This species resembles in aspect *Peniophora cremea* and *P. mutata*, and its hyphae are similarly coarse and loosely arranged but both cystidia and gloeocystidia are lacking. The incrusted zone at the base of the subhymenium is about 30  $\mu$  thick and very characteristic. Each hypha assumes incrustation upon entering this zone, has position parallel to the other hyphae, and is devoid of incrustation beyond the zone.

Specimens examined:

Porto Rico: Rio Piedras, Palo Seco, La Isabell Grove, J. A. Stevenson, 3523, type (in Mo. Bot. Gard. Herb., 6635).

28. C. lacteum Fries, Epicr. 560. 1838; Hym. Eur. 649. 1874; Sacc. Syll. Fung. 6: 610. 1888; Massee, Linn. Soc. Bot. Jour. 27: 132. 1890.—Not *C. lacteum* of Bresadola, v. Höhnel & Litschauer, nor probably of Bourdot & Galzin, and Rea.

Thelephora lactea Fries, Syst. Myc. 1: 452. 1821; Elenchus Fung. 1: 205. 1828.—Corticium pellicula (Fr. ?) Karsten, Soc. pro Fauna et Fl. Fenn. Meddel. 11: 5. 1885.

Type: in Fries Herb.—the specimen determined by E. Fries. Authentic specimen in better condition in Kew Herb.—the cream-colored fructification collected by Lbd., Svex. Soderm, Oct.

Fructifications effused, thin, membranaceous, tender, small pieces separable, becoming cream-colored to cinnamon-buff in the herbarium, even, more or less cracked, the margin whitish, fibrillose; in section 150–300  $\mu$  thick, not colored, with the hyphae densely and longitudinally arranged along the substratum and then curving upward to the hymenium,  $2\frac{1}{2}-4$   $\mu$  in diameter, incrusted in the subhymenial region, occasionally nodose-septate; no gloeocystidia nor vesicular bodies; spores hyaline, even, subglobose, about  $5-6\frac{1}{2}\times 5-6$   $\mu$ , pointed at the base.

Fructifications 3-8 cm. long, 2-5 cm. wide.

On decaying wood and limbs of coniferous and frondose species and on the ground. In Europe and in northern United States and Canada from Massachusetts westward to the Pacific states. May to November. Occasional.

It has been necessary to depart from the conflicting concepts of C. lacteum, and base the species on the presumably oldest existing specimen collected and determined by Fries and preserved in his herbarium under the name of Thelephora lactea. Other and more recent specimens were referred by Fries to Corticium lacteum, the genus Corticium not being used by Fries until the publication of his 'Epicrisis.' These more recent specimens are of various species as might be expected, for the exact methods of the present day in the study of resupinate Hymenomycetes were not then used, and it is probable that these later specimens have caused the confusion in current concepts of C. lacteum. It is fortunate that one of these later specimens, named by Fries, agrees with the original specimen, is in better condition than the original specimen, and is preserved in Kew Herbarium where it is convenient for comparison. C. lacteum, as understood from these specimens, belongs in a group of species of similar aspect having globose spores about 6 \(\mu\) in diameter. The other members of this group are C. radiosum, and C. abeuns. C. pelliculare has the same aspect as the others named but its spores are not globose. one knows any one of the above group of species the other species should be readily recognized as they are found, for C. lacteum has rather coarse, loosely arranged, more or less granule-incrusted hyphae, and lacks gloeocystidia and vesicular bodies: C. abeuns has wholly immersed gloeocystidia of the usual kind; and C. radiosum has vesicular organs which are at first like those of C. polyonium but become much more inflated and with highly attenuated wall, and finally perhaps are shown only by vesicular spaces between the massed hyphae.

Specimens examined:

Sweden: type, under the name *Thelephora lactea* (in Herb. Fries); Svex. Söderm., *Lindblad*, determined by E. Fries as *Corticium lacteum* (in Kew Herb.); Stockholm, *L. Romell*, 114, 179, 327.

Finland: Mustiala, P. Karsten, comm. by Karsten under the name Corticium pellicula Fr.?

France: Fautrey, determination as Corticium lacteum approved by Patouillard for Lloyd, comm. by C. G. Lloyd, 4368.

Canada: J. Macoun, 29, and an unnumbered specimen from Ellis Herb., comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 44640); Ironsides, J. Macoun, 286 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61348); Lower St. Lawrence Valley, J. Macoun, 45, 47, 90; Ottawa, J. Macoun, 57, and 165 and 349 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 56052, 55921).

Vermont: Middlebury, E. A. Burt, 2 gatherings.

Massachusetts: Magnolia, W. G. Farlow.

New York: Albany, H. D. House & J. Rubinger, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 7462); Altamont, E. A. Burt; Freeville, G. F. Atkinson, 2586; Hague, C. H. Peck, 11; Ithaca, G. F. Atkinson, 2870, 14100; Sandlake, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55748); Warrensburg, C. H. Peck, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 55976).

Pennsylvania: Carbondale, E. A. Burt.

Alabama: Montgomery, R. P. Burke, 220 (in Mo. Bot. Gard. Herb., 57094).

Ohio: Cincinnati, A. P. Morgan, comm. by Lloyd Herb., 2617.

Michigan: Ann Arbor, C. H. Kauffman, 24, 38 (in Mo. Bot. Gard. Herb., 17172, 18617); New Richmond, C. H. Kauffman, 25, 33, 42 (in Mo. Bot. Gard. Herb., 17035, 20030, 22870).

Wisconsin: Blue Mounds, A. O. Stucki, 37.

Illinois: Peoria, C. J. Humphrey, 1990 (in Mo. Bot. Gard. Herb., 17518).

Idaho: Bonanza, G. G. Hedgcock, comm. by C. J. Humphrey, 2557, in part; Coolin, J. R. Weir, 11574 (in Mo. Bot. Gard. Herb., 63302).

Colorado: Uncompaghre National Forest, G. G. Hedgcock, comm. by C. J. Humphrey, 2546.

British Columbia: Sidney, J. Macoun, 84 (in Mo. Bot. Gard. Herb., 55346).

Washington: Chiquash Mountains, W. N. Suksdorf, 842; Seattle, W. A. Murrill, 151, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 55727).

California: Massack, A. S. Rhoads, 21 (in Mo. Bot. Gard. Herb., 56990).

29. C. subgiganteum Berkeley, Grevillea 2: 3. 1873; Sacc. Syll. Fung. 6: 632. 1888; Lyman, Boston Soc. Nat. Hist. Proc. 33: 151. pl. 18, f. 2-21, pl. 26, f. 137. 1907.

Peniophora subgigantea (Berk.) Massee, Linn. Soc. Bot. Jour. 25: 142. 1889.—Michenera artocreas Berk. & Curtis, Linn. Soc. Bot. Jour. 10: 333. 1868; Sacc. Syll. Fung. 6: 653. 1888; Patouillard, Soc. Myc. Fr. Bul. 7: 42. pl. 4, f. 1-5. 1891; Essai Taxon. 67. 1900; Peirce, Torr. Bot. Club Bul. 17: 305. pl. 110, f. k-n. 1890; Lyman, Boston Soc. Nat. Hist. Proc. 33: 157. pl. 18, f. 6-21, pl. 26, f. 137. 1907.—An Corticium gilvidum Bresadola, Ann. Myc. 18: 46. 1920?

Type: in Kew Herb. and Farlow Herb.

Basidiosporic stage broadly effused, adnate, thick, membranaceous, separable in small pieces when moist, drying light buff to light ochraceous-buff, even, glabrous, not cracked, the margin whitish, sometimes buff when old; in section 500–1000  $\mu$  thick, not colored, with the hyphae densely interwoven, about  $2-2\frac{1}{2}\,\mu$  in diameter, not incrusted, not nodose-septate; no gloeocystidia; paraphyses with pointed tips; basidia large with 4 sterigmata usually; basidiospores hyaline, even, globose or subglobose,  $14-19~\mu$  in diameter or  $14-19~\times~12-16~\mu$ .

Chlamydosporic or *Michenera* fructifications disk-shaped, concave, drying snuff-brown, cracked, the margin acute, thick, white on its elevated side; in section 1–2 mm. thick, composed of a thick basal layer of densely interwoven hyphae about 2  $\mu$  in diameter which terminate in sporiferous ends and branches densely crowded together in the concave layer at surface of the fructifications; sporophores consist of each a single chlamydospore terminating in a slender, flexuous, tapering terminal appendage up to 10–50  $\mu$  long; chlamydospores ovoid, even,  $12–20 \times 10–15 \mu$ .

Basidiosporic fructifications 2-15 cm. long, 1-4 cm. wide; *Michenera* fructifications 6-8 mm. in diameter.

On bark of dead limbs of Acer rubrum, Magnolia, and Lirio-dendron. In swamps in the Atlantic states from Canada to Cuba. July to February. Occasional.

Fructifications of the perfect stage bear some resemblance in general aspect to those of *C. portentosum* but are readily distinguished by the much larger spores. When growing on the

same twigs the perfect fructifications occur normally on the under side of the twigs with the imperfect ones opposite on the upper side.

Specimens examined:

Exsiccati: Ell. & Ev., N. Am. Fungi, 3102, under the name Corticium ochroleucum var. resupinatum.

Canada: Quebec, Hull, J. Macoun, 149; Ontario, Ottawa, J. Macoun, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 55802).

Maine: Kittery Point, R. Thaxter, comm. by G. R. Lyman.

New Hampshire: Chocorua, W. G. Farlow (in Mo. Bot. Gard. Herb., 55580); North Conway, comm. by L. O. Overholts, 5062 (in Mo. Bot. Gard. Herb., 56354).

Vermont: Middlebury, C. G. Lloyd, 10623 (in Mo. Bot. Gard. Herb., 44639).

Connecticut: near Moosup River, J. L. Sheldon, comm. by C. J. Humphrey, 2526 (in Mo. Bot. Gard. Herb., 18559).

New York: Karner, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55782).

New Jersey: Newfield, J. B. Ellis, comm. by Lloyd Herb., 1442, by Farlow Herb. (in Mo. Bot. Gard. Herb., 55584), and in Ell. & Ev., N. Am. Fungi, 3102.

Virginia: Clarendon, W. H. Long, 12715 (in Mo. Bot. Gard. Herb., 55060).

North Carolina: Transylvania County, W. A. Murrill & H. D. House, 423, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 56586).

South Carolina: Aiken, H. W. Ravenel, 1669, type (in Kew Herb. and Farlow Herb.).

Alabama: Auburn, G. F. Atkinson, 2364.

Cuba: C. Wright, type of Michenera artocreas (in Farlow Herb.).

C. ceraceum Berk. & Rav. in Ravenel, Fungi Car. Exs.
 1855, without description; Massee, Linn. Soc. Bot. Jour.
 150. 1890; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 785. text f. 6. 1907.

Corticium molle Berk. & Curtis, Linn. Soc. Bot. Jour. 10: 336. 1868; Grevillea 1: 180. 1873.—Not Corticium molle Fries.—C. armeniacum Sacc. Syll. Fung. 6: 637. 1888.

Type: type distribution in Ravenel, Fungi Car. 3: 29.

Fructifications broadly effused, ceraceous-fleshy, drying membranaceous, small pieces separable when moistened, becoming cinnamon-buff to army-brown in the herbarium, even, shining, not cracking, the margin paler, narrow, with hyphae interwoven; in structure  $100-400~\mu$  thick, not colored, composed of erect, densely interwoven, agglutinate, thick-walled hyphae  $2\frac{1}{2}-3~\mu$  in diameter, not incrusted, rarely, if at all, nodose-septate; no gloeocystidia; spores hyaline, even, flattened on one side,  $10-16~\times 4\frac{1}{2}-7~\mu$ .

Fructifications 1-10 cm. long, 1-3 cm. wide; sometimes confluent over areas up to 1 m. long.

On decaying trunks of frondose species. New Jersey to Mexico, in the West Indies, and in South Africa. Throughout the year. Uncommon.

C. ceraceum varies in the thickness of its fructifications which are usually cinnamon to ochraceous-orange in color and sometimes become very large. The spores are so very large that they afford a good distinctive character but are most likely to be found in crushed preparations of the hymenium.

Specimens examined:

Exsicatti: Ellis, N. Am. Fungi, 607; Ravenel, Fungi Am., 453; Fungi Car. 3: 29, type distribution.

New Jersey: Newfield, J. B. Ellis, comm. by Lloyd Herb.

Virginia: Woodstock, C. L. Shear, 1193.

North Carolina: Biltmore Estate, W. A. Murrill (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61351).

South Carolina: H. W. Ravenel, in Ellis, N. Am. Fungi, 607, and type in Ravenel, Fungi Car. 3: 29; Aiken, H. W. Ravenel, in Ravenel, Fungi Am., 453; Black Rock, H. W. Ravenel, 1261 (in Curtis Herb.).

Alabama: Montgomery, R. P. Burke, 133 (in Mo. Bot. Gard. Herb., 10951).

Louisiana: Lafayette County, A. B. Langlois, 1467; St. Martinville, A. B. Langlois, 41, comm. by Lloyd Herb., and 2709, and G.

Mexico: Orizaba, W. A. & E. L. Murrill, 789, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54615).

Cuba: C. Wright, type of Corticium molle B. & C. (in Curtis Herb., 202); Alto Cedro, Earle & Murrill, comm. by N. Y. Bot. Gard. Herb.

Africa: locality not given, P. A. van der Bijl, 13 (in Mo. Bot. Gard. Herb., 58810).

### 31. C. Bambusae Burt, n. sp.

Type: in Burt Herb., and Farlow Herb.

Fructifications small, becoming confluent, effused, adnate, very thin, tender, small pieces separable, cartridge-buff, even, not shining, somewhat cracked, the margin free in some places; in section 80–120  $\mu$  thick, not colored, with the hyphae about  $2\frac{1}{2}\,\mu$  in diameter, not incrusted, not nodose-septate, arranged longitudinally along the substratum and sending out lateral branches to form the hymenium; no gloeocystidia; basidia simple, 40  $\times$  10  $\mu$ , with 4 sterigmata; spores hyaline, even, 14–18  $\times$  8–9  $\mu$ , pointed at both ends, copious.

Fructifications 1-3 mm. in diameter, becoming confluent over an area 4 cm. long,  $1-1\frac{1}{2}$  cm. wide.

On bamboo. West Indies and Venezuela. Very common.

The small, cartridge-buff fructifications clustered together and becoming confluent over the hard cortex of culms of bamboo and the unusually large spores are good distinctive characters for recognition of this species.

Specimens examined:

Trinidad: Maravel, R. Thaxter, type, comm. by W. G. Farlow, 19.

32. C. cremoricolor Berk. & Curtis, Grevillea 1: 180. 1873; Sacc. Syll. Fung. 6: 615. 1888.—Massee, Linn. Soc. Bot. Jour. 27: 133. 1890 (spelled cremicolor).

Type: in Kew Herb. and Farlow Herb., labelled Corticium cremicolor B. & C.

Fructifications broadly effused, rather thick, membranaceous, small pieces separable when moistened, becoming cream-colored and pinkish buff to wood-brown in the herbarium, cracking into areolae 2–3 mm. in diameter and with a distinctly radial arrangement of the principal cracks frequently, more or less colliculose with broad, slightly elevated granules, the margin narrow, fibril-

lose, sometimes radiate; in section 200–800  $\mu$  thick, not colored, with hyphae somewhat longitudinally interwoven and then ascending to a compact hymenium, 2–3  $\mu$  in diameter, rarely larger, not incrusted but mixed with more or less mineral matter; no gloeocystidia; spores hyaline, even, flattened on one side, 8–12  $\times$  5–8  $\mu$ .

Fructifications 2-10 cm. long, 1½-3 cm. wide, often confluent. On bark of decaying *Quercus* and other frondose species. Throughout Canada and the United States. April to December. Frequent.

C. cremoricolor is so similar to C. hydnans in aspect that the much larger spores of C. cremoricolor afford the best character for separation of these two species. C. cremoricolor is less tubercular, however, thicker, and usually with cracks radiating from the center of the fructification. C. anthracophilum Bourd. is closely related in structure.

Specimens examined:

Canada: J. Macoun, 19.

Massachusetts: Cambridge, L. M. Underwood, 1001 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 57340).

New York: Ithaca, H. S. Jackson, comm. by Cornell Univ. Herb., 14391; Onondaga Valley, L. M. Underwood (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61388).

New Jersey: Belleplain, C. L. Shear, 1247, Newfield, J. B. Ellis, comm. by Farlow Herb. (in Mo. Bot. Gard. Herb., 44636).

Pennsylvania: Ohio Pyle, W. A. Murrill, 1076, 1133 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61586, 61579); Reitz Gap, L. O. Overholts, 4633 (in Mo. Bot. Gard. Herb., 56118); State College, L. O. Overholts & C. R. Orton, comm. by L. O. Overholts, 4723 (in Mo. Bot. Gard. Herb., 56115); Trexlertown, W. Herbst, comm. by Lloyd Herb., 2227; Whitehaven, G. F. Atkinson, 8654.

Maryland: Takoma Park, C. L. Shear, 1075.

District of Columbia: Washington, C. L. Shear, 1240, 1259.

Florida: W. W. Calkins, comm. by Farlow Herb. (in Mo. Bot. Gard. Herb., 44633).

Alabama: Peters, type (in Kew Herb., and Curtis Herb., 5205). Texas: Houston, H. W. Ravenel, 271, comm. by U. S. Dept. Agr.

- Herb. This is the *Corticium lactescens* of Cooke's Fungi of Texas; Quitman, W. H. Long, 12092 (in Mo. Bot. Gard. Herb., 55047).
- Ohio: C. G. Lloyd, 3821, comm. by Farlow Herb., 166 (in Mo. Bot. Gard. Herb., 55261), and 3821 and 3907.
- Indiana: Scottsburg, J. R. Weir, 376 (in Mo. Bot. Gard. Herb., 17186).
- Illinois: Christopher, C. J. Humphrey, 2092 (in Mo. Bot. Gard. Herb., 21145); Lombard, E. T. & S. A. Harper, 952.
- Michigan: Ann Arbor, C. H. Kauffman, 27; New Richmond, Demmon, comm. by A. H. W. Povah, 6 (in Mo. Bot. Gard. Herb., 20198), and C. H. Kauffman, 29 (in Mo. Bot. Gard. Herb., 20304).
- Wisconsin: Madison, M. C. Jensen, and another comm. by C. J. Humphrey, 2439 (in Mo. Bot. Gard. Herb., 43839 and 22376, respectively); Stevens Point, C. J. Humphrey, 1802 (in Mo. Bot. Gard. Herb., 17910).
- Minnesota: Univ. Minn. Myc. Herb., comm. by E. L. Jensen, 8 (in Mo. Bot. Gard. Herb., 10565).
- Missouri: Bismarck, L. O. Overholts (in Mo. Bot. Gard. Herb., 58322).
- British Columbia: Sidney, J. Macoun, 12 (in Mo. Bot. Gard. Herb., 5730).
- New Mexico: Cienega Springs, W. H. Long, 21596 (in Mo. Bot. Gard. Herb., 55120); Cloudcroft, W. H. Long, 19665, 19523 (in Mo. Bot. Gard. Herb., 55044, 55045); Tyom Canyon, W. H. Long, 21895 (in Mo. Bot. Gard. Herb., 55119); Tyom Exp. Sta., W. H. Long, 21877 (in Mo. Bot. Gard. Herb., 55118).
- 33. C. confluens Fries, Epicr. 564. 1838; Hym. Eur. 655. 1874; Berkeley, Outl. Brit. Fung. 276. 1860; Sacc. Syll. Fung. 6: 626. 1888; Massee, Linn. Soc. Bot. Jour. 27: 133. 1890; Bresadola, I. R. Accad. Agiati Atti III. 3: 112. 1897; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 252. 1911; Rea, Brit. Basid. 679. 1922.
- Thelephora confluens Fries, Syst. Myc. 1: 447. 1821.—Corticium confluens var. subcalceum Karsten, Rev. Myc. 10:74. 1888. Fructifications effused, rather thick, waxy-membranaceous,

small pieces separable when moistened, whitish to cartridge-buff and light pinkish cinnamon in the herbarium, even, with few cracks, the margin indeterminate, thinning out; in section 200–500  $\mu$  thick, not colored, composed of ascending, densely interwoven and agglutinate, thin-walled hyphae  $2\frac{1}{2}-3$   $\mu$  in diameter, not incrusted, occasionally nodose-septate; no gloeocystidia; spores hyaline, even, ovoid,  $5-9 \times 3\frac{1}{2}-6$   $\mu$ , copious.

Fructifications 2-8 cm. long, 1-3 cm. wide.

On bark of fallen decaying limbs of *Betula*, *Alnus*, *Salix*, and other frondose species. In Europe, from Newfoundland to Louisiana and westward to Manitoba and Washington, in Mexico, the West Indies, Japan, and South Africa. April to December. Common.

C. confluens may be recognized among our species by its occurrence on frondose bark in closely adnate fructifications with somewhat the aspect of pale Peniophora incarnata but of different structure, which is distinctive by not being stratose and by having the hyphae agglutinate, and by the presence of large spores. The authentic specimen from Karsten of C. confluens var. subcalceum has spores  $9 \times 6 \mu$  and does not have cystidia, differing in both respects from the statement by Bresadola in Ann. Myc. 1: 102. 1903.

Specimens examined:

Sweden: L. Romell, 80, 81, 82, 83, 84.

Finland: Mustiala, authentic specimen, perhaps part of type of Corticium confluens var. subcalceum Karst. from Karsten.

Germany: Lengerich, W. Brinkmann, Westfälische Pilze, 13 (in Mo. Bot. Gard. Herb., 63430).

Austria: Tirol, Hall, V. Litschauer; Stubai, V. Litschauer.

Italy: G. Bresadola.

Newfoundland: Bay of Islands, A. C. Waghorne, 983 (in Mo. Bot. Gard. Herb., 63747).

Canada: Lower St. Lawrence Valley, J. Macoun, 65.

Ontario: Eastman's Springs, J. Macoun, 532; Ottawa, J. Macoun, 29; Woodstock, E. Bartholomew, 6713 (in Mo. Bot. Gard. Herb., 57041).

New Hampshire: Camp, Ellis R., *Underwood & C.*, 22 (in N. Y. Bot. Gard. Herb., Burt Herb., and Mo. Bot. Gard. Herb., 61585); Chocorua, W. G. Farlow.

Vermont: Middlebury, E. A. Burt, 2 gatherings.

New England: W. G. Farlow.

Massachusetts: Waverly, G. R. Lyman, 164.

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57446, 57472, 57670, 59680); Altamont, E. A. Burt; East Galway, E. A. Burt; Hudson Falls, S. H. Burnham, 48 (in Mo. Bot. Gard. Herb., 54465); Ithaca, C. H. Kauffman, C. O. Smith, Van Hook, comm. by G. F. Atkinson, 14384, 8045, and 8048, respectively; Karner, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 54374, 55206); New York, Class in Mycology (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61346); North Elba, C. H. Peck, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 56111); Oneida, H. D. House, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 59705); Seventh Lake, Hamilton County, H. E. Stork, 2 (in Mo. Bot. Gard. Herb., 56639); West Park, New York City, F. S. Earle, 1596 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61425); West Troy, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55781).

New Jersey: Belleplain, C. L. Shear, 1255.

Pennsylvania: Carbondale, E. A. Burt; Germantown, E. A. Burt; State College, L. O. Overholts, 2620 (in Mo. Bot. Gard. Herb., 20278).

Maryland: Silver Springs, D. G. Fairchild, comm. by U. S. Dept. Agr. Herb.

District of Columbia: Takoma Park, C. L. Shear, 1354; Washington, C. L. Shear, 1238, in part.

Florida: Daytona, R. A. Harper, 9 (in Mo. Bot. Gard. Herb., 54536).

Alabama: Auburn, F. S. Earle & C. F. Baker, and 43 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61558).

Louisiana: Baton Rouge, Edgerton & Humphrey, comm. by C. J. Humphrey, 5729, 5733; St. Martinville, A. B. Langlois, i, dh, and 472 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 614788), and 1761a, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 42600).

Illinois: Duquoin, C. J. Humphrey, 1309, 1394 (in Mo. Bot. Gard. Herb., 10324, 10352); Riverside, E. T. & S. A. Harper, 675.

Wisconsin: Blue Mounds, Miss Stucki, 12, 13.

Iowa: Ames, H. H. Hume, 3 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61583); Fort Dodge, O. M. Oleson, 438 (in Mo. Bot. Gard. Herb., 44077).

Manitoba: Winnipeg, G. R. Bisby, 62 (in Mo. Bot. Gard. Herb., 57898).

Washington: Puyallup, C. J. Humphrey, 7649.

Porto Rico: Campo Alegre, J. A. Stevenson, 6585 (in Mo. Bot. Gard. Herb., 55078).

Jamaica: Troy, A. E. Wight, 420, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 14558).

Mexico: Guernavaca, W. A. & E. L. Murrill, 541, 543, 548, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54558, 54559, 54560).

Japan: Prov. Shinauo, A. Yasuda, 133 (in Mo. Bot. Gard. Herb., 62060).

Africa: Natal, Pietermaritzburg, P. A. van der Bijl, 583 (in Mo. Bot. Gard. Herb., 69371).

34. Coniophora corrugis Burt, Mo. Bot. Gard. Ann. 13: 310. 1926.

This species occurs on living trees, logs and dead limbs of conifers in forests of the Rocky Mountain region and from British Columbia to Arizona in the Pacific states. The fructifications are somewhat coriaceous, loosely attached to the substratum, and vinaceous in color. The spores in most specimens are colorless, even,  $6-10 \times 4-7 \,\mu$ , not copious—fully mature and colored in only one of the specimens received during 14 years.

35. C. laetum (Karst.) Bresadola, Ann. Myc. 1: 94. 1903; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1552. 1906; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 237. 1911.

Hyphoderma laetum Karsten, Rev. Myc. 11: 206. 1889; Sacc. Syll. Fung. 10: 530. 1892.—Corticium hypnophilum Karsten, Rev. Myc. 12: 126. 1890; Sacc. Syll. Fung. 9: 234. 1891.

Fructifications effused, thin, membranaceous-waxy, soft, small pieces separable when moist, orange-pink to rose color, fading in the herbarium to cartridge-buff, even, not cracked, the margin

thinning out, somewhat arachnoid; in section 100–200  $\mu$  thick, not colored, composed of interwoven, hyaline hyphae 5–8  $\mu$  in diameter, not incrusted, no clamp connections found; no gloeocystidia; spores hyaline, even, 6–12  $\times$  4–8  $\mu$ .

Fructifications 5 mm.-2 cm. long, 5-10 mm. wide.

On living mosses and on bark of dead Alnus and Betula. In Europe and in New York, Michigan, and North Dakota.

This species may be recognized by bright rose color when fresh, occurrence on living moss and dead alders, large spores, coarse hyphae, and absence of gloeocystidia. The three American specimens cited below seem referable to C. laetum except that their hyphae are more numerous and of smaller diameter—4-6  $\mu$ —than those of the European specimens with which compared. Peniophora aurantiaca has much the same aspect and occurs on Alnus also but has gloeocystidia and cystidia.

Specimens examined:

Sweden: L. Romell, 145.

Finland: Mustiala, authentic specimen of C. hypnophilum from Karsten.

Italy: specimen on Alnus of C. laetum collected and determined by Bresadola.

New York: Karner, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 44708).

Michigan: Isle Royale, Allen & Stuntz, 42, comm. by Univ. Wis. Herb.

North Dakota: Brenckle, comm. by V. Litschauer, 2.

36. C. roseum Persoon, Roemer Neues Mag. Bot. 1: 111. 1794; Fries, Epicr. 560. 1838; Hym. Eur. 650. 1874; Berkeley, Outl. Brit. Fung. 273. 1860; Sacc. Syll. Fung. 6: 611. 1888; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 233. 1911; Coker, Elisha Mitchell Scientif. Soc. Jour. 36: 171. pl. 33, f. 3-5. 1921; Rea, Brit. Basid. 673. 1922.

Thelephora rosea Persoon, Syn. Fung. 575. 1801; Myc. Eur. 1:131. 1822; Fries, Syst. Myc. 1:451. 1821; Elench. Fung. 1:203. 1828.—Corticium roseolum Massee, Linn. Soc. Bot. Jour. 27:140. pl. 6, f. 2. 1890.—C. polygonoides Karsten, Soc. pro Fauna et Fl. Fenn. Meddel. 6:12. 1881; Sacc. Syll. Fung. 6:

638. 1888; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 232. 1911.—Lyomyces polygonoides Karsten, Finska Vet.-Soc. Bidrag Natur. och Folk 48: 419. 1889.—Aleurodiscus roseus (Pers.) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1568. 1906.

Fructification effused, rather thick, adnate, somewhat membranaceous, small pieces separable when moist, drying pinkish buff to buff-pink, pruinose, finally cracked, the margin whitish, more or less byssoid; in section 200–280  $\mu$  thick, with the hymenial layer perhaps slightly colored, 2-layered, with the basal layer composed of longitudinally arranged, densely interwoven hyphae 3–3½  $\mu$  in diameter, not incrusted, the hymenial layer composed of erect hyphae, basidia, and slender, slightly brownish, short-branched paraphyses; no gloeocystidia; basidia at first exceeded by the paraphyses, finally protruding; spores hyaline, even, 6–12  $\times$  4½–8  $\mu$ .

Fructifications sometimes 2-3 mm. in diameter and becoming laterally confluent, more usually 1-10 cm. long, 1-3 cm. wide.

On bark and wood of logs and branches of frondose species such as *Populus*, *Betula*, *Alnus*, *Acer*, *Carya*, *Ulmus*, etc., rarely on coniferous wood. In Europe, from Canada to Alabama, westward to Manitoba and Washington, in New Mexico and Mexico, and in Japan. Throughout the year. Common.

C. roseum is well named, for its pale rose-color is distinctive and is confirmed, when sections are examined, by the slender, slightly brownish, short-branched organs which are probably paraphyses but have seemed to me when in young vigorous condition to have the branches tipped by very minute spherical bodies. C. polygonoides is the early stage with the paraphyses exceeding the young basidia.

Specimens examined:

Exsiccati: Ell. & Ev., Fungi Col., 609, under the name Corticium incarnatum; de Thümen, Myc. Univ., 2012.

Sweden: L. Romell, 47, 127, 146; Stockholm, L. Romell, 147.

Finland: Mustiala, P. A. Karsten, in de Thümen, Myc. Univ., 2012. and authentic specimen of Lyomyces polygonoides.

Austria: Stubai, Tirol, V. Litschauer.

Italy: Trient, G. Bresadola.

England: Apthorpe, Norths, 12, type of C. roseolum (in Kew Herb.).

Canada: J. Macoun, 85.

Ontario: London, J. Dearness, D1078c (in Mo. Bot. Gard. Herb., 18666); Ottawa, J. Macoun, 135, 451.

Maine: Freeport, O. O. Stover, comm. by P. L. Ricker.

Vermont: Middlebury, E. A. Burt, 3 gatherings; Ripton, E. A. Burt, 2 gatherings; Smugglers' Notch, Mt. Mansfield, E. A. Burt.

Massachusetts: Newton, W. G. Farlow; Willow Brook, H. Webster, comm. by Boston Myc. Club Herb., E.; Waverly, G. R. Lyman, 120, 164.

New York: Alcove, C. L. Shear, 1204, 1313; Altamont, E. A. Burt; Ithaca, G. F. Atkinson, 2120, and H. S. Jackson, comm. by Cornell Univ. Herb., 14389; Minnewaska, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55980); Orient, R. Latham, 223 (in Mo. Bot. Gard. Herb., 44226); Poughkeepsie, R. C. Poppey, in Gerard Herb. (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61559); Syracuse, L. M. Underwood, 18 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 44312); White Plains, L. M. Underwood (in N. Y. Bot. Gard. Herb., 61410).

New Jersey: Newfield, J. B. Ellis, in Ell. & Ev., Fungi Col., 609. Pennsylvania: Center Hall, E. West, comm. by L. O. Overholts, 3659 (in Mo. Bot. Gard. Herb., 54700); State College, J. Ellis, comm. by L. O. Overholts, 5207 (in Mo. Bot. Gard. Herb., 56360).

District of Columbia: Takoma Park, C. L. Shear, 953.

North Carolina: W. C. Coker, 4703 (in Mo. Bot. Gard. Herb., 57424).

Alabama: Auburn, C. F. Baker (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 61397, and Burt Herb.); Montgomery, R. P. Burke, 2, in part, 160, 305 (in Mo. Bot. Gard. Herb., 22073, 44961, 57195).

Ohio: College Hill, Aiken, comm. by Lloyd Herb., 2341; Linwood, C. G. Lloyd, 1870; Preston, C. G. Lloyd, 1561.

Indiana: Indianapolis, *H. von Schrenk* (in Mo. Bot. Gard. Herb., 19805).

Illinois: Cairo, E. Bartholomew, 9234.

Minnesota: Brickton, C. J. Humphrey, 1124 (in Mo. Bot. Gard. Herb., 10276).

Iowa: Decorah, E. W. D. Holway.

Missouri: Creve Coeur Lake, F. P. McWhorter (in Mo. Bot. Gard. Herb., 57334).

Montana: Monarch, J. R. Weir (in Mo. Bot. Gard. Herb., 20736). Idaho: J. R. Weir, 366 (in Mo. Bot. Gard. Herb., 15165).

Manitoba: River Park, A. H. R. Buller, 873 (in Mo. Bot. Gard. Herb., 58994); Stony Mountain, A. H. R. Buller, 897 (in Mo. Bot. Gard. Herb., 58989); Winnipeg, A. H. R. Buller, 936 (in Mo. Bot. Gard. Herb., 59025).

Washington: Bingen, W. N. Suksdorf, 685, 720; Columbia River, W. Klickitat County, W. N. Suksdorf, 106.

New Mexico: Mogollon, G. G. Hedgcock & W. H. Long, comm. by C. J. Humphrey, 2540 (in Mo. Bot. Gard. Herb., 21660).

Mexico: Parral, Chihuahua, E. O. Mathews, 4 (in Mo. Bot. Gard. Herb., 44419).

Japan: Sendai, A. Yasuda, 60 (in Mo. Bot. Gard. Herb., 56144).

37. C. salmonicolor Berk. & Broome, Linn. Soc. Bot. Jour. 14: 71. 1873; Sacc. Syll. Fung. 6: 620. 1888; Massee, Linn. Soc. Bot. Jour. 27: 122. 1890; Petch, Phys. and Dis. of Hevea brasiliensis, 209. 1911; Rorer, Trinidad Dept. Agr. Bul. 15<sup>3</sup>: 1. f. 1, 2. 1917; Lee & Yates, Philippine Jour. Sci. 14: 657. pl. 1-7. 1919.

Necator decretus Massee, Kew Bul. Misc. Inf. 1898: 119. 1898; Sacc. Syll. Fung. 16: 1094. 1902.—Corticium javanicum Zimmermann, Centralbl. f. Bakt. Abt. 2, 7: 103. text f. 3. 1901.—C. Zimmermanni Sacc. & Syd. in Sacc. Syll. Fung. 16: 1117. 1902; 17: 169. 1905.

Type: in Kew Herb.

Fructifications broadly effused, thin, adnate, membranaceous-soft, separable when moist, pale ochraceous buff to orange-pink when fresh, fading in the herbarium to pale olive-buff and cartridge-buff, pulverulent, even, cracking a little in drying, the margin thinning out; in section 100–200  $\mu$  thick, composed of hyphae running longitudinally over the substratum and bearing

a broad layer of suberect, branching, loosely interwoven hyphae 4–7  $\mu$  in diameter, not incrusted, not nodose-septate; no gloeocystidia; basidiospores hyaline, even, 9–12  $\times$  6–8  $\mu$ . The conidia of the imperfect Necator stage are catenulate, 14–18  $\times$  7–8  $\mu$ , according to Massee.

Fructifications 2-20 cm. long, 1-3 cm. wide.

Parasitic on bark of branches 1-3 cm. in diameter and young trees of *Cacao*, *Citrus*, *Hevea*, *Amherstia*, tea and coffee plants in tropical regions, and on *Ficus* and pear and apple shoots in Florida and Louisiana. In West Indies, Philippine Islands, East Indies, and Ceylon.

C. salmonicolor is a species very destructive to important economic species of shrubs and trees, causing the Pink Disease where the climate is warm and moist for sufficiently long periods that the mycelium can run over the bark of young shoots and penetrate into the deeper tissues. Its parasitic occurrence on living woody plants, bright color, coarse hyphae, and large spores render it easy to recognize in tropical regions.

Specimens examined:

Florida: Gainesville, J. Matz (in Mo. Bot. Gard. Herb., 44822, 54934).

Louisiana: Baton Rouge, C. W. Edgerton, 702, 990a.

Porto Rico: Bayamon, J. A. Stevenson, 2827 (in Mo. Bot. Gard. Herb., 9689); Pueblo Vigo, J. A. Stevenson, 5436 (in Mo. Bot. Gard. Herb., 7820); Trujillo Alto, J. A. Stevenson, 3819, and W. C. Drier, comm. by J. A. Stevenson, 6770 (in Mo. Bot. Gard. Herb., 9059 and 55054, respectively).

Dominica: W. Norwell, comm. by J. B. Rorer (in Mo. Bot. Gard. Herb., 18560).

Trinidad: J. B. Rorer (in Mo. Bot. Gard. Herb., 20429); Guaico, J. B. Rorer, four gatherings (in Mo. Bot. Gard. Herb., 14023, 17934, 20295, 44770); Port of Spain, J. B. Rorer (in Mo. Bot. Gard. Herb., 9008).

Ceylon: a portion of 3 authentic specimens determined by Berkeley in Kew Herb. (in Mo. Bot. Gard. Herb., 8891), T. Petch, comm. by Kew Herb. (in Mo. Bot. Gard. Herb., 8890); Peradeniya, T. Petch, 8640 (in Mo. Bot. Gard. Herb., 56245).

#### 38. C. spretum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, adnate, rather thick, somewhat coriaceous, cinnamon-buff in the herbarium, even, not shining, in drying cracking to the substratum into polygonal masses about 1 mm. in diameter, the margin similar, narrow, entire; in section 200–300  $\mu$  thick, colored like the hymenium, composed of ascending, densely interwoven, thin-walled hyphae  $3-3\frac{1}{2}\mu$  in diameter, not incrusted, not nodose-septate; no gloeocystidia; slender paraphyses about  $1\mu$  in diameter, with short branches near the tips, are present between the basidia; spores hyaline, even,  $8-10\times5-6\mu$ .

Fructifications probably large, for received in fragments up to 5 cm. long, 2 cm. wide.

On decorticated wood of a decaying stump of  $Fraxinus\ oregona$ . Washington. September.

C. spretum has conspicuous fructifications resembling Hymenochaete spreta in aspect. The deeply cracked fructifications cinnamon-buff externally and throughout, large spores, slender paraphyses, and occurrence on ash stumps should enable the species to be recognized confidently.

Specimens examined:

Washington: Bingen, W. N. Suksdorf, 962, type.

39. C. rubropallens (Schw.) Massee, Linn. Soc. Bot. Jour. 27: 145. 1890.

Thelephora rubropallens Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 168. 1832.—Stereum rubropallens (Schw.) Cooke, Grevillea 20: 35. 1891; Sacc. Syll. Fung. 11: 121. 1895.—Not C. rubropallens Bresadola, Ann. Myc. 1: 97. 1903, nor Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 258. 1911.

Type: in Schweinitz Herb. and probably in Farlow Herb. and Kew Herb.

"T. effusa, indeterminatim effigurata, ambitu marginibus latissimis albis; versus centrum subroseo-incarnata, crebre sporidifera aut pulverulenta. Pelliculam efficit ex arcte intertextis filis. Ulnarem longitudinem explet.

"Longe lateque effusa in corticibus et lignis Bethlehem."

In section 100–150  $\mu$  thick, not colored, with the hyphae suberect, branched, rather loosely interwoven, about  $2\frac{1}{2}-3~\mu$  in diameter, not incrusted but bearing imbedded crystalline matter, with very slender, colorless, hair-like paraphyses protruding beyond the basidia and, in my opinion, with short branches near the tips; no gloeocystidia; spores noted by Massee as  $8-9\times3~\mu$ , and by Cooke as  $6-7\times3~\mu$ , none found in my preparations of the type.

I regret that a Corticium on Fagus, Ripton, Vermont, Nov. 4, 1896, which I misdetermined as C. rubropallens, relying too largely on general aspect and coloration in comparison with the type, and communicated to Bresadola, Romell, and Karsten under that name, should have led both Bresadola and Bourdot into error concerning C. rubropallens. The names of those specimens should be changed to C. roseopallens Burt, as described in the present work.

C. rubropallens belongs in the group of species with C. rubro-canum, C. albido-carneum, and C. Atkinsonii. Each species of this group lacks gloeocystidia and has the very slender and numerous paraphyses protruding beyond the basidia and masking the latter. The only recent gathering which I can now refer to C. rubropallens on the basis of agreement in internal structure is now white in herbarium condition and doubtful therefore. Its few spores are  $9-10 \times 4 \mu$ .

Specimens examined:

Pennsylvania: Bethlehem, Schweinitz, type (in Schweinitz Herb.). Alabama: Montgomery, R. P. Burke, 118 (in Mo. Bot. Gard. Herb., 19557).

40. C. rubrocanum de Thümen, Myc. Univ., 409, with description. 1876; Torr. Bot. Club Bul. 6: 95. 1876; Sacc. Syll. Fung. 6: 632. 1888.

Type: type distribution in de Thümen, Myc. Univ., 409.

Fructifications broadly effused, thin, adnate, membranaceous, small pieces separable when moist, becoming tilleul-buff in the herbarium, hoary, glabrous, finally cracking at the center into polygonal masses 1–2 to a mm., the margin determinate or indeterminate and thinning out, of the same color; in section

100–150  $\mu$  thick, not colored or only very slightly in the subhymenium, with the hyphae longitudinally and densely interwoven next to the substratum, then becoming erect, bushybranched in the hymenial layer, short-celled, of irregular outline, about 3–3½  $\mu$  in diameter, not incrusted but with some imbedded crystalline matter; paraphyses slightly brownish below, protruding beyond the basidia as very slender hairs about ½–1  $\mu$  in diameter with short lateral branches; no gloeocystidia; the only spore found is hyaline, even,  $9 \times 3\frac{1}{2}$   $\mu$  but may not belong.

Fructifications 2-10 cm. long, 1-2 cm. wide.

On fallen twigs of *Quercus coccinea*. New Jersey to Louisiana. November to April. Not common.

P. rubrocanum is distinguished by its occurrence in thin, hoary, nearly white fructifications with a tint of pink on small fallen branches of oak, and by the absence of gloeocystidia and the presence of delicate hair-like paraphyses in the hymenial surface. Spore collections should be made to determine the spore dimensions, for the spores have not been retained well in any specimen examined. It is probable that C. rubrocanum will be found to be a synonym of C. rubropallens when the type of the latter can be studied more critically than by me twenty-six years ago.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 22; de Thümen, Myc. Univ., 409, type.

New Jersey: Newfield, J. B. Ellis, in Ellis, N. Am. Fungi, 22, in de Thümen, Myc. Univ., 409, and (in Mo. Bot. Gard. Herb., 4846, 44638).

South Carolina: H. W. Ravenel, 377, comm. under the name C. Auberianum by N. Y. Bot. Gard. Herb.

Alabama: Montgomery, R. P. Burke, 105 (in Mo. Bot. Gard. Herb., 11280).

Louisiana: Natchitoches, G. D. Harris, comm. by Cornell Univ. Herb., 5111; St. Martinville, A. B. Langlois, 1933

# 41. C. cultum Burt, n. sp.

Type: in Burt Herb.

Fructifications usually a thin, whitish, cottony mycelium along the sides of tunnels of a bark beetle but sometimes bearing a hymenium and in those places effused, small, thin, closely adnate, somewhat membranaceous-fleshy, ivory-yellow when growing, fading to white in the herbarium, even, not cracked, the margin continuous with the sterile mycelium; in section  $100-150~\mu$  thick, not colored, composed of suberect, branching, densely arranged and somewhat interwoven hyphae  $3-3\frac{1}{2}~\mu$  in diameter, short-celled, occasionally nodose-septate; no gloeocystidia; basidia simple, cylindric,  $27~\times~3\frac{1}{2}-4\frac{1}{2}~\mu$ , with 4 knob-shaped sterigmata; spores hyaline, even,  $6-8~\times~3\frac{1}{2}-4\frac{1}{2}~\mu$ , copious; some imbedded spores present.

Fructifications 5-10 mm. long, 1-2 mm. wide.

In thick bark of coniferous logs on side walls of tunnels made by a bark-boring beetle. Idaho probably.

C. cultum is one of the species which should be considered in connection with the fungous flora of burrows of bark-boring insects. The term "ambrosia fungi" has been used for some other fungi growing in such places. The type specimen of C. cultum is scanty but well fruited. The species has not been received from any source as growing on the exterior of bark or wood.

Specimens examined:

Idaho: probably Idaho but only general locality stated, J. R. Weir, comm. by W. G. Farlow, type (in Mo. Bot. Gard. Herb., 44655).

### 42. C. rubellum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, adnate, thin, somewhat membranaceous, small pieces separable when moist, vinaceous-fawn, becoming wood-brown in the herbarium, even, not waxy, the margin thinning out; in section 120–500  $\mu$  thick, not colored when thin but somewhat colored in thick fructifications and then stratose, with the hyphae arranged longitudinally and crowded together parallel with the substratum in each stratum, more loosely interwoven towards the hymenium,  $2\frac{1}{2}$ –3  $\mu$  in diameter, not incrusted, rarely nodose-septate; no gloeocystidia; spores copious, hyaline, even, 6–9  $\times$  5–6  $\mu$ , flattened on one side, with a small apiculus on the flattened side near the base.

Fructifications 5-10 cm. long, 1-5 cm. wide.

On decorticated wood of dead *Vitis* and on decaying bark of *Quercus Gambelii* and *Tilia*. Florida, Illinois, Colorado, and Manitoba. July to October.

C. rubellum differs from C. rubicundum in becoming finally stratose and somewhat colored, having larger and more subglobose spores, and occurring on dead grape vines, oak, and basswood. The Florida specimen lacks spores and may be incorrectly referred here. C. confluens has similar spores.

Specimens examined:

Florida: New Smyrna, W. A. Murrill, 27, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 62081).

Illinois: Glencoe, E. T. & S. A. Harper, 941, type; River Forest, E. T. & S. A. Harper, 661.

Colorado: Deer Creek Park, E. Bartholomew, 9149, 9150.

Manitoba: Winnipeg, A. H. R. Buller, comm. by G. R. Bisby, 724 (in Mo. Bot. Gard. Herb., 58987).

### 43. C. hydnans (Schw.) Burt, n. comb.

Radulum hydnans Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 164. 1832; Sacc. Syll. Fung. 11: 112. 1895.—Corticium colliculosum Berk. & Curtis, Grevillea 2: 3. 1873; Peck, N. Y. State Mus. Rept. 28: 52. 1876; Sacc. Syll. Fung. 6: 618. 1888: Massee, Linn. Soc. Bot. Jour. 27: 134. 1890.

Type: in Farlow Herb. and probably in Schweinitz Herb.

Fructifications long and widely effused, adnate, thin, membranaceous, small pieces separable when moistened, pinkish buff to cinnamon-buff in the herbarium, becoming more or less colliculose or somewhat tuberculate, cracking into polygonal masses 1–2 mm. in diameter, the margin whitish, with hyphae interwoven; in structure 100–300  $\mu$  thick, not colored, with the hyphae longitudinally arranged next the substratum and then ascending and interwoven to the hymenium, 2–3  $\mu$  in diameter, not incrusted; no gloeocystidia; spores hyaline, even, 5–8  $\times$  2½–3½  $\mu$ .

Fructifications 1-10 cm. long, 1-3 cm. wide.

On decaying frondose limbs on the ground. Canada to Texas and westward to Washington and British Columbia. April to November. Occasional.

C. hydnans is intermediate between Corticium and Radulum with granules rather too broad at base, too little elevated and too convex to be a typical Radulum in configuration, and yet always leading one to search for more raduloid teeth. It is well named as C. hydnans or by its later name C. colliculosum. It may be distinguished from Radulum orbiculare in doubtful cases by its lack of gloeocystidia.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 329 and 717 b, the latter under the name *Corticium subgiganteum*; Ravenel, Fungi Am., 126, 227, both under the name *Corticium calceum*; de Thümen, Myc. Univ., 605.

Canada: Gaspé, J. Macoun, 530.

Ontario: London, J. Dearness, 1178 (in Mo. Bot. Gard. Herb., 18773).

New Hampshire: North Conway, A. S. Rhoads, 7 (in Mo. Bot. Gard. Herb., 56893).

Vermont: Middlebury, E. A. Burt.

Massachusetts: Sprague, 96, type of Corticium colliculosum (in Curtis Herb., 5297).

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 14834); Alcove, C. L. Shear, 1011, 1212, 1219; East Galway, E. A. Burt; Grand View, H. von Schrenk (in Mo. Bot. Gard. Herb., 42817); Ithaca, H. S. Jackson, comm. by Cornell Univ. Herb., 14390, and W. H. Long (in Mo. Bot. Gard. Herb., 62987); New Baltimore, C. H. Peck, comm. by N. Y. State Mus. Herb., T 30 (in Mo. Bot. Gard. Herb., 56071); Trenton Falls, C. H. Peck, comm. by N. Y. State Mus. Herb., T 9 (in Mo. Bot. Gard. Herb., 54572).

New Jersey: Newfield, J. B. Ellis, and (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61636) and in Ellis, N. Am. Fungi, 717 b, and de Thümen, Myc. Univ., 605.

Pennsylvania: Bethlehem, Schweinitz, type of Radulum hydnans (in Schweinitz Herb. and Farlow Herb.); Center County, C. R. Orton, comm. by L. O. Overholts, 2940 (in Mo. Bot. Gard. Herb., 8265); State College, L. O. Overholts, 3040 (in Mo. Bot. Gard. Herb., 5689); Trexlertown, W. Herbst, 3.

Maryland: Rock Creek, C. L. Shear, 1046.

District of Columbia: Washington, C. L. Shear, 1261.

North Carolina: Biltmore Estate, W. A. Murrill (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61382).

Georgia: Darien, H. W. Ravenel, in Ravenel, Fungi Am. 227.

Florida: Gainesville, H. W. Ravenel, in Ravenel, Fungi Am., 126.

Louisiana: Baton Rouge, Edgerton & Humphrey, comm. by C. J. Humphrey, 5642; St. Martinville, A. B. Langlois, comm. by U. S. Dept. Agr. Herb.

Texas: Lindheimer, 40 (in Mo. Bot. Gard. Herb., 4819).

West Virginia: Paw Paw, C. L. Shear, 1175.

Kentucky: Crittenden, C. G. Lloyd, 2365, 3118.

Ohio: Cincinnati, comm. by Lloyd Herb., 2792; Loveland, D. L. James, comm. by U. S. Dept. Agr. Herb.

Illinois: Glen Ellyn, E. T. & S. A. Harper, 955; River Forest, E. T. & S. A. Harper, 734.

Michigan: Ann Arbor, C. H. Kauffman, 48 (in Mo. Bot. Gard. Herb., 8083); Gogebic County, E. A. Bessey, 248 (in Mo. Bot. Gard. Herb., 56613).

Missouri: Grandin, H. von Schrenk (in Mo. Bot. Gard. Herb., 43021).

Nebraska: Long Pine, C. L. Shear, 1065.

British Columbia: Yoho Valley, J. Macoun, 6.

Washington: Bellingham, J. R. Weir, 545 (in Mo. Bot. Gard. Herb., 5899).

California: Santa Catalina Island, L. W. Nuttall, 402, in part (in Mo. Bot. Gard. Herb., 57614).

# 44. C. rubicundum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, rather thick, membranaceous, loosely attached, separable, drying buff-pink to light vinaceous-cinnamon, slightly tubercular, pruinose, the margin radiating, whitish; in section 200–500  $\mu$  thick, not colored, with a hymenial layer 60  $\mu$  thick borne on a broad layer reaching to the substratum and composed of interwoven, thick-walled, hyaline hyphae 3–4  $\mu$  in diameter, not incrusted, occasionally nodose-septate; no gloeocystidia; basidia 4-spored; spores hyaline, even, 4–7  $\times$  3–4½  $\mu$ , copious.

Fructifications 6-8 cm. long, 2-5 cm. wide.

On bark of logs of *Tsuga canadensis*, *Picea* and *Pinus*. Canada, Colorado and Washington. September.

C. rubicundum has large, sheet-like, loosely attached fructifications with somewhat the aspect of those of *Peniophora* velutina but lacking cystidia. The thick, membranaceous, loosely attached fructification is suggestive of a resupinate *Stereum* but I have seen no *Stereum* of which this may be the resupinate stage. The occurrence on hemlock bark should help in identifying future gatherings.

Specimens examined:

Canada: Lake Rosseau, Ontario, E. T. & S. A. Harper, 637, type. Colorado: near Mancos, G. G. Hedgcock, comm. by C. J. Humphrey, 2560.

Washington: Mt. Paddo, W. N. Suksdorf, 735, 736.

## 45. C. granulatum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, thin, closely adnate, central portions fawn-color, becoming wood-brown in the herbarium, dull rather than shining, with some scattered, small granules, not cracked, the margin fimbriate, fading from ochroleucous to whitish; in section 120–240  $\mu$  thick, not colored, with a narrow incrusted zone, the hyphae densely interwoven, 3  $\mu$  in diameter, somewhat incrusted, not nodose-septate; no gloeocystidia; basidia protruding slightly when mature, with 4 sterigmata; spores hyaline, even,  $4\text{--}5 \times 2\text{--}3~\mu$ .

Fructifications 3-5 cm. long, 1-2 cm. wide.

On very rotten wood of *Populus trichocarpa*. Idaho. September and October.

This species should be readily recognized by its color when fresh, somewhat granular hymenium, and occurrence on decaying poplar wood. The incrustation of the hyphae is a good available character for separation from *C. subceraceum* and *C. deflectens*.

Specimens examined:

Idaho: Priest River, J. R. Weir, 33, type, and 106.

46. C. illaqueatum Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 238. 1911.

Type: authentic specimens in Burt Herb.

Fructifications effused, adnate, membranaceous-thin, loosely attached to the substratum, small pieces separable when moist, becoming cream-buff in the herbarium, even, not cracked regularly, the margin somewhat arachnoid; in section 150–300  $\mu$  thick, not colored, composed of loosely interwoven, thin-walled hyphae 3–4  $\mu$  in diameter, nodose-septate, with some incrustation next to the substratum; no gloeocystidia; spores hyaline, even,  $4\frac{1}{2}$ –6  $\times$  3  $\mu$ , borne 4 to a basidium.

Fructifications 1-3 cm. long,  $\frac{1}{2}$ -1 $\frac{1}{2}$  cm. wide.

On bark of decaying Castanea and other frondose species. France and Louisiana. September to January.

C. illaqueatum has color somewhat like that of C. ceraceum and C. hydnans but is loosely attached to the substratum and has smaller spores than the former and does not crack in drying like the latter.

Specimens examined:

France: Aveyron, H. Bourdot, 16063, and M. Galzin, 12684, 12689, 15107, comm. by H. Bourdot, 18548, 16092, 12623.

Louisiana: St. Martinville, A. B. Langlois, 203.

# 47. C. Rosae Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, adnate, rather thick, membranaceous, separable, drying Rood's brown, ceraceous, even, contracting in drying and cracking through the hymenial layer into rectangular masses 2–4 mm. in diameter and showing the thick, white, cottony subiculum in the crevices, the margin white, cottony; in section 400–600  $\mu$  thick, not colored, with the hyphae about 3  $\mu$  in diameter, more or less incrusted in the middle region, not nodose-septate, densely crowded together and arranged longitudinally in a broad layer along the substratum, then ascending obliquely and becoming densely interwoven in a thick hymenial layer; no gloeocystidia; spores hyaline, even, 4–7  $\times$  2½–3  $\mu$  as seen attached to the basidia.

Fructifications received in fragments  $2-2\frac{1}{2}$  cm. long, 1 cm. wide—broken off on three sides.

On bark of dead wild rose—Rosa sp. British Columbia. February.

C. Rosae has thick fructifications which are conspicuous by their reddish brown color and prominent white margin. The occurrence on wild rose bushes should aid in recognition of the species. The loose attachment to the substratum by a broad layer of longitudinally arranged hyphae is suggestive of the genus Stereum but the specimens do not have the margin reflexed in the least degree; I know of no Stereum of which this may be the resupinate stage.

Specimens examined:

British Columbia: Sidney, J. Macoun, 275, type (in Mo. Bot. Gard. Herb., 63772) and another specimen of the same number comm. by J. Dearness (in Mo. Bot. Gard. Herb., 63773).

48. C. apiculatum Bresadola, Mycologia 17: 68. 1925.

C. areolatum Bresadola, Mycologia 17: 68. 1925.

Type: in Weir Herb.

Fructifications broadly effused, thin, membranaceous, tender, small portions separable when moistened, between ivory-yellow and cream color, even, contracting in drying and cracking into angular masses about 1 mm. in diameter more or less completely separated by fissures which show the floccose subiculum along their sides, the margin thinning out, fibrillose; in section 90–130  $\mu$  thick, not colored, composed of loosely interwoven, thin-walled hyphae  $2\frac{1}{2}-4\frac{1}{2}\mu$  in diameter, with an occasional incrusting granule, occasionally nodose-septate; no gloeocystidia; spores hyaline, even,  $4\frac{1}{2}-5\times2\frac{1}{2}-3\mu$ .

Fructifications 2-5 cm. long,  $1\frac{1}{2}$ -3 cm. wide.

On decaying branches of Alnus tenuifolia. Alabama to Idaho, and British Columbia to Mexico. October and December.

C. apiculatum belongs in the C. lacteum group of species. It should be recognized in its region by occurrence on Alnus, cream color, and small, somewhat elliptical spores. C. areolatum has a fructification with the areolate masses separated from one another by rather wide fissures but of same color as type of C. apiculatum, spores the same size, and fructification separable to the same degree—certainly not closely adnate.

Specimens examined:

Alabama: Montgomery, R. P. Burke, 199, 202, 671 (in Mo. Bot. Gard. Herb., 57075, 57078, 63102).

Missouri: near St. Louis, L. O. Overholts, 3167 (in Mo. Bot. Gard. Herb., 5711).

Idaho: Priest River, J. R. Weir, 23304, type (in Weir Herb.), and 23387, type of C. areolatum (in Weir Herb.).

British Columbia: Sidney, J. Macoun, 33 (in Mo. Bot. Gard. Herb., 6767).

Washington: Seattle, W. A. Murrill, 131, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 55742).

Mexico: Jalapa, W. A. & E. L. Murrill, 123, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 10748).

#### 49. C. subceraceum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, adnate, membranaceous, separable when moist, tawny to hazel in the herbarium, even or with some small obtuse granules, waxy, not cracking, the margin somewhat fimbriate, whitish; in section 200–300  $\mu$  thick, not colored, 2-layered, the layer next to the substratum thick, composed of loosely arranged, suberect hyaline hyphae not incrusted, not nodose-septate, mostly  $4-4\frac{1}{2}\mu$  in diameter but with a few up to  $6\mu$ , the hymenial layer dense, thin, undulating; no gloeocystidia; spores hyaline, even,  $4-4\frac{1}{2}\times 2-2\frac{1}{2}\mu$ .

Fructifications 3-8 cm. long, 1-3 cm. wide.

On wood and bark of fallen frondose limbs, rarely on pine. July to October. Pennsylvania to Alabama and westward to Illinois. Infrequent.

C. subceraceum resembles in general aspect C. ceraceum but has small spores. This species should be compared with Grandinia mucida when the problem of the latter is being solved; the only European specimen of G. mucida which I have studied was shared with me by Bresadola and is distinct, having aspect of the illustration in Fries, Icones Hym., pl. 195, f. 3.

Specimens examined:

Pennsylvania: Trexlertown, W. Herbst, 76, type, and an unnumbered specimen, both received under the name Corticium laeve of Herbst, Fung. Fl. Lehigh Valley.

Maryland: Takoma Park, C. L. Shear, 1275.

District of Columbia: W. A. Murrill, 1446 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61491).

North Carolina: Salem, Schweinitz, the Thelephora aurantia of Schweinitz, Fungi Car. and Thelephora (Grandinia) mucida of Schweinitz, Syn. N. Am. Fungi, 708 (in Schweinitz Herb.).

Alabama: Montgomery, R. P. Burke, 191 (in Mo. Bot. Gard. Herb., 57070).

Kentucky: Crittenden, C. G. Lloyd, 1684, 3123.

Ohio: C. G. Lloyd, 4177, 4179; Cincinnati, C. G. Lloyd, 4496; Madisonville, C. G. Lloyd, 0171.

Illinois: Cerro Gordo, L. O. Overholts, 3121 (in Mo. Bot. Gard. Herb., 5715); River Forest, E. T. & S. A. Harper, 658.

**50.** C. roseo-pallens Burt in Lyman, Boston Soc. Nat. Hist. Proc. **33**: 173. pl. 20, f. 56-73. 1907.

Type: in Burt Herb.

Fructifications broadly effused, thin, adnate, membranaceous, tender, small pieces separable when moist, flesh-pink when fresh, fading to ivory-yellow in the herbarium, at first with the hymenium interrupted, at length continuous, waxy, even, the margin thinning out, with the hyphae interwoven; in section 100–200  $\mu$  thick, not colored, with the hyphae suberect, interwoven, more loosely arranged near the substratum,  $3-3\frac{1}{2}$   $\mu$  in diameter, not incrusted, occasionally nodose-septate; no gloeocystidia; basidia 4-spored; spores pale rose when first collected, fading to white, even, cylindric, slightly curved,  $4-5 \times 1\frac{1}{2}-2$   $\mu$ .

Fructifications 3-12 cm. long, 2-6 cm. wide.

On bark and wood of decaying logs of Fagus, Populus, Quercus, etc. Maine to Louisiana and in Missouri. October. Occasional.

This species may be recognized by its broadly effused, thin, flesh-pink or pale rosy salmon fructifications, fading upon drying to nearly white and by the small allantoid spores. In his discussion of a portion from my type, comm. to Bresadola under the name C. rubropallens, Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 258. 1911, regard their C. subtestaceum as a synonym and C. incrustans v. Höhn. & Litsch. as scarcely distinct. I have not seen specimens of the latter species and those of the former, communicated to me by Bourdot, are hardly convincing.

Specimens examined:

- Maine: Kittery Point, R. Thaxter, comm. by W. G. Farlow, 7 (in Mo. Bot. Gard. Herb., 55291).
- Vermont: Grand View Mountain, E. A. Burt; Middlebury, E. A. Burt; Ripton, E. A. Burt, type; Weybridge, E. A. Burt.
- Massachusetts: Stony Brook, G. R. Lyman, 142; Waverly, G. R. Lyman, 142.
- New York: Albany, H. D. House, 14.170 and H. D. House & J. Rubinger (in Mo. Bot. Gard. Herb., 44721, 8732); Ithaca, G. F. Atkinson, 2559a; Sylvan Beach, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 9089).
- Louisiana: Lafayette, A. B. Langlois, 1764, comm. by W. G. Farlow.
- Missouri: Creve Coeur, B. M. Duggar (in Mo. Bot. Gard. Herb., 44821).
- 51. C. ochraceum Fries, Epicr. 563. 1838; Hym. Eur. 652. 1874; Berkeley, Outl. Brit. Fung. 275. 1860; Sacc. Syll. Fung. 6: 624. 1888; Bresadola, Fungi Trid. 2: 60. pl. 170, f. 1. 1898; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 256. 1911; Rea, Brit. Basid. 680. 1922.

Thelephora calcea var. argillacea Fries, Elench. Fung. 1: 215. 1828.

Type: in Fries Herb.

Fructifications broadly effused, closely adnate, rather thick, becoming pinkish buff to wood-brown in the herbarium, waxy, even or somewhat papillose, contracting in drying and cracking to the substratum into rectangular masses about  $\frac{1}{2}-1$  mm. in diameter, and showing sides of the fissures composed of firm, dense, agglutinate structure, the margin at first whitish, soon concolorous, thinning out; in section  $300-500\,\mu$  thick, becoming somewhat zonate or stratose, not colored, composed of erect hyphae densely crowded, interwoven, and so closely glued together that the deeply staining lumen is the distinguishable part; gloeocystidia, if present at all, so similar to the hyphae in form and diameter that there is no indication of them except in aqueous mounts; spores hyaline, even,  $4-6\times21/2-31/2$   $\mu$ .

Fructifications 3-10 cm. long, 1-4 cm. wide.

On decorticated and sometimes charred limbs on the ground of

Pinus Strobus and other conifers. In Europe and in Vermont, Alabama, Idaho, and Washington. September and October. Rare in North America.

C. ochraceum somewhat resembles in general aspect C. lactescens and is, in my opinion, related to the latter by hyphae in barely the beginning of differentiation into gloeocystidia. C. ochraceum of American plant lists is based on misdetermined specimens.

Specimens examined:

Sweden: Femsjö, E. Fries, type (in Fries Herb.); North Sweden, L. Romell, 403; Smöland, E. Fries, authentic specimen of Corticium calceum var. argillaceum (in Fries Herb.).

Austria: Innsbruck, Tirol, V. Litschauer.

Italy: on Abies excelsa in Alps Mts., G. Bresadola.

Vermont: Middlebury, E. A. Burt.

Alabama: Montgomery, R. P. Burke, 606 (in Mo. Bot. Gard. Herb., 57471).

Montana: Rexford, E. E. Hubert, comm. by J. R. Weir, 12017 (in Mo. Bot. Gard. Herb., 63373).

Idaho: Priest River, J. R. Weir, 59.

Washington: Hoquiam, C. J. Humphrey, 6373; Seattle, C. J. Humphrey, 6454, and W. A. Murrill, 135, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 55737).

# 52. C. furfuraceum Bresadola, Mycologia 17: 69. 1925. Type: in Weir Herb.

Fructifications broadly effused, closely adnate, thin, furfuraceous, ivory-yellow to pinkish buff in the herbarium, becoming somewhat cracked, the margin thinning out, pruinate; in section  $60-140~\mu$  thick, not colored, composed of suberect, thin-walled hyphae about  $3~\mu$  in diameter, somewhat collapsed and irregular in outline, indistinct, not incrusted; no gloeocystidia nor conducting organs; spores hyaline, even,  $4-5\frac{1}{2}~\times~2\frac{1}{2}~\mu$ .

Fructifications more than 10 cm. long, for broken off at both ends, 6 cm. wide.

On decaying wood of logs of Abies grandis, Pinus monticola, P. contorta, P. ponderosa, and Larix occidentalis. Montana, Idaho, Washington, and British Columbia. August and September. Probably common.

C. furfuraceum may be recognized on the substrata given by the very thin, closely adnate fructifications of ivory-yellow to pinkish buff color, which crack slightly by contraction in drying and have small spores.

Specimens examined:

Montana: Evaro, J. R. Weir, 439 (in Mo. Bot. Gard. Herb., 63714); Missoula, J. R. Weir, 401, 409 (in Mo. Bot. Gard. Herb., 11316, 63717).

Idaho: Coolin, J. R. Weir, 17211, type, 16764 and 16927 (in Weir Herb.).

British Columbia: Kootenai Mountains, near Salmo, J. R. Weir, 481, 501, 526 (in Mo. Bot. Gard. Herb., 63725, 63716, 63715). Washington: Kalama, C. J. Humphrey, 6225.

53. C. lividum Persoon, Obs. Myc. 1: 38. 1796; Fries, Epicr. 563. 1838; Hym. Eur. 652. 1874; Berkeley, Outl. Brit. Fung. 275. 1860; Sacc. Syll. Fung. 6: 623. 1888; Massee, Linn. Soc. Bot. Jour. 27: 152. 1890; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 253. 1911; Rea, Brit. Basid. 680. 1922.

Thelephora livida Persoon, Myc. Eur. 1: 148. 1822; Fries, Syst. Myc. 1: 447. 1821; Elench. Fung. 1: 218. 1828.—Phlebia livida (Pers.) Bresadola, I. R. Accad. Agiati Atti III. 3: 105. 1897.—Grandinia ocellata Fries, Epicr. 527. 1838; Hym. Eur. 626. 1874; Sacc. Syll. Fung. 6: 501. 1888.—An Corticium hepaticum Berk. & Curtis, Grevillea 1: 180. 1873?

Fructifications broadly effused, agglutinated, sometimes becoming rather thick, somewhat waxy-gelatinous, not separable, varied in color, gray or tinged reddish or bluish, becoming pale smoke gray, cinnamon-buff, and raisin-black in the herbarium, pruinose, even or sometimes radiately wrinkled or tuberculate by aggregations of imbedded crystalline matter, the margin thinning out, similar or whitish; in section 100–500  $\mu$  thick, not colored usually, rarely slightly brownish, composed of densely interwoven, suberect hyphae about 2–3  $\mu$  in diameter, with the walls gelatinously modified and glued together; no gloeocystidia; spores hyaline, even, 3–5  $\times$  1½–2–2  $\mu$ .

Fructifications 2-10 cm. long, 1-5 cm. wide.

On rotting logs, usually decorticated, of coniferous species,

more rarely on frondose logs. In Europe, Canada to Texas, and westward to British Columbia and California, and in Venezuela and East Indies. June to December. Probably common.

C. lividum may be recognized by its livid fructifications of gray, reddish, or bluish tinge and of somewhat gelatinous consistency, somewhat suggestive of those of Peniophora gigantea in aspect but destitute of cystidia. C. hepaticum seems to me referable to C. lividum but I need to study the type again in the feature of the slightly reflexed margin, which I now suspect may be that of a different species overrun by C. lividum. Since the tubercules of the Grandinia ocellata form are due to heaps of imbedded crystals, it has seemed to taxonomists that this species is a Corticium rather than a Grandinia.

Specimens examined:

Sweden: Femsjö, L. Romell, 214, E. A. Burt, 3 gatherings, L. Romell, comm. by Bresadola.

Austria: Steiermark, N. Rechinger, comm. & det. by V. Litschauer; Tirol, V. Litschauer.

Hungary: Kmet, comm. by Bresadola.

Italy: G. Bresadola.

England: Mulgrave Woods, E. M. Wakefield (in Mo. Bot. Gard. Herb., 57115).

Canada: J. Macoun, 94, and 350, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 8269); J. Dearness (in Mo. Bot. Gard. Herb., 56797); Ottawa, J. Macoun, 2, 46, 53.

New Hampshire: Chocorua, W. G. Farlow.

Vermont: Middlebury, E. A. Burt.

New York: Ampersand, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 56102); Catskill Mts., C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55792).

Pennsylvania: State College, L. O. Overholts, 3425 (in Mo. Bot. Gard. Herb., 54471).

Maryland: Takoma Park, C. L. Shear, 1269.

Louisiana: St. Martinville, A. B. Langlois, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 44693), and comm. by Ellis Herb. (in Burt Herb.).

Texas: Silsbee, W. H. Long, 21227 (in Mo. Bot. Gard. Herb., 55127).

Wisconsin: Lake Geneva, E. T. & S. A. Harper, 839.

Montana: Anaconda, E. E. Hubert, comm. by J. R. Weir, 12007 (in Mo. Bot. Gard. Herb., 63367); Como, E. E. Hubert, comm. by J. R. Weir, 11958 (in Mo. Bot. Gard. Herb., 63315); Evaro, J. R. Weir, 421 (in Mo. Bot. Gard. Herb., 14764); Kalispell, E. E. Hubert, comm. by J. R. Weir, 11972 (in Mo. Bot. Gard. Herb., 63333); Libby, E. E. Hubert, comm. by J. R. Weir, 11347, 11360, 12041 (in Mo. Bot. Gard. Herb., 63701, 63702, 63391); Missoula, E. E. Hubert, comm. by J. R. Weir, 11981 (in Mo. Bot. Gard. Herb., 63334); Radnor, E. E. Hubert, comm. by J. R. Weir, 11645 (in Mo. Bot. Gard. Herb., 63707).

Idaho: Coeur d'Alene, E. E. Hubert, comm. by J. R. Weir, 11993 (in Mo. Bot. Gard. Herb., 63356); Priest River, J. R. Weir, 6364 (in Mo. Bot. Gard. Herb., 58373), and 13, 76, 84; Santa, E. E. Hubert, comm. by J. R. Weir, 11755, 12003, 12042 (in Mo. Bot. Gard. Herb., 63313, 63365, 63392).

British Columbia: Kootenai Mts. near Salmo, J. R. Weir, 527 (in Mo. Bot. Gard. Herb., 20903); Sidney, J. Macoun, 85, 380 (in Mo. Bot. Gard. Herb., 63693, 63764); Vancouver Island, J. Macoun, comm. by J. Dearness, V 85 (in Mo. Bot. Gard. Herb., 22729).

Washington: Kalama, C. J. Humphrey, 6138.

Oregon: Philomath, S. M. Zeller, 2159 (in Mo. Bot. Gard. Herb., 58774).

California: Requa, R. Kelly, comm. by A. S. Rhoads, 16 (in Mo. Bot. Gard. Herb., 56985).

Venezuela: La Guayra, A. F. Blakeslee, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 55294).

East Indies: Batavia, *Rick*, comm. by Bresadola under the name *Phlebia livida* (Pers.) Bres.

# 54. C. Overholtsii Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, thin, somewhat membranaceous, at first between pale salmon and pale grayish vinaceous, becoming tilleul-buff in the herbarium, even, pruinose, not cracked, the margin thinning out, somewhat fimbriate; in section 160  $\mu$  thick, not colored, composed of suberect, densely inter-

woven, conglutinate hyphae up to 3  $\mu$  in diameter, not incrusted, with wall gelatinously modified, the outline not well defined; no gloeocystidia; spores hyaline, even, 5-6  $\times$  2-2½  $\mu$ , copious.

Fructifications  $1\frac{1}{2}$ -3 cm. long, 1-2 cm. wide.

On thick bark of dead Pinus rigida. Pennsylvania. October.

C. Overholtsii has the livid color of C. vinaceo-scabens but nothing else in common with that species. In structural details it is related to C. lividum but does not have the appearance of dried cartilage or resin, characteristic of all specimens of the latter known to me.

Specimens examined:

Pennsylvania: Reitz Gap, L. O. Overholts, 4656, type (in Mo. Bot. Gard. Herb., 57155).

## 55. C. Pseudotsugae Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, not at all separable, light buff in the herbarium, even, not shining, not cracked, the margin similar, thinning out, pulverulent; in section 45–55  $\mu$  thick, not appreciably colored, composed of densely interwoven hyphae about  $1\frac{1}{2}-2$   $\mu$  in diameter, not incrusted, conglutinate; no gloeocystidia; basidia with 4 sterigmata; spores hyaline, even,  $3-5 \times 2-3$   $\mu$ .

Fructifications 5-8 cm. long, 1-2 cm. wide.

On decorticated, decaying wood of *Pseudotsuga taxifolia* and *Tsuga canadensis*. New York and Idaho. August to November.

C. Pseudotsuga is almost exactly the avellaneous color of Saccardo's 'Chromotaxia.' This color, occurrence on hemlock, and very thin fructifications are the most available characters for recognition of the species.

Specimens examined:

New York: Freeville, G. F. Atkinson, 2627.

Idaho: Sandpoint, E. E. Hubert, comm. by J. R. Weir, 11617, type (in Mo. Bot. Gard. Herb., 63305).

56. C. confine Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 260. 1911.

Type: authentic specimen in Burt Herb.

Fructifications broadly effused, thin, closely adnate, pale pinkish buff to pale olive-buff in the herbarium, not shining, hypochnoid, rimose-granular into areas or granules about 2–3 to a mm., the margin thinning out, byssoid; in section 75–150  $\mu$  thick, not colored, composed of erect, thin-walled, hyaline hyphae  $2\frac{1}{2}-3$   $\mu$  in diameter, of irregular outline, collapsing, nodose-septate; no gloeocystidia; spores hyaline, even, 3–5  $\times$   $2\frac{1}{2}$   $\mu$ , copious.

Fructifications 4-10 cm. long, 2-4 cm. wide.

On decaying frondose wood. France and Vermont. May to August.

This species is related to *Grandinia* by its granular aspect but the granules seem to have originated so largely from the cracking of the fructification to the substratum that I concur in the inclusion in *Corticium*. It has a more hypochnoid surface than *C. scutellare*.

Specimens examined:

France: Allier, H. Bourdot, 16064, 16065.

Vermont: Middlebury, Battell Ledge, E. A. Burt.

# 57. C. analogum (B. & G.) Burt, n. comb.

Gloeocystidium analogum Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 366. 1913.

Type: authentic specimen in Burt Herb.

Fructifications broadly effused, thick, adnate, fleshy-membranaceous, small pieces separable when moistened, becoming maize-yellow to chamois in the herbarium, somewhat colliculose, not cracked or but little cracked, not shining, the margin thinning out; in section 500–1000  $\mu$  thick, colored like the hymenium, becoming zonate or somewhat stratose, composed of hyphae 3–4  $\mu$  in diameter, densely interwoven, conglutinate and poorly defined, of great numbers of imbedded spores and gloeocystidia; gloeocystidia immersed in many zones or layers, 40–80  $\times$  6–8  $\mu$ , becoming dissolved by potassium hydrate solution; imbedded spores subglobose, 5–6  $\times$  5  $\mu$ , minutely rough, slightly colored in the deeper portions of the fructification, hyaline at the surface of the hymenium; spores on basidia not demonstrated.

Fructifications in fragments up to 8 cm. long, 3 cm. wide.

On decaying wood of *Quercus* and *Fraxinus* in France, of *Quercus* in Maine, and of *Populus trichocarpa* in Idaho. July to October. Probably rare.

C. analogum has general aspect and color of C. galactinum and C. portentosum and structure related to that of C. effuscatum. The thick, stratose fructifications, containing great numbers of imbedded spores and gloeocystidia, afford good additional distinctive characters. The Maine specimens are doubtfully referred here as a young first-stratum stage.

Specimens examined:

France: Aveyron, A. Galzin, 12435, authentic specimen, comm.

by H. Bourdot, 16164.

Maine: Kittery Point, R. Thaxter & E. A. Burt.

Idaho: Priest River, J. R. Weir, 25.

58. C. effuscatum Cooke & Ellis, Grevillea 9: 103. 1881;
Sacc. Syll. Fung. 6: 633. 1888; Massee, Linn. Soc. Bot. Jour.
27: 142. 1890; Lyman, Boston Soc. Nat. Hist. Proc. 33: 176. pl. 21, f. 74-95, pl. 22, f. 96-105. 1907.

Type: in Kew Herb.

Fructifications broadly effused, rather thick, membranaceous, small pieces separable when moistened, honey-yellow to russet when fresh, fading to cream-buff in the herbarium, even, pulverulent, the margin thinning out; in section 200–500  $\mu$  thick, composed of very densely arranged, suberect, interwoven hyphae about 2  $\mu$  in diameter, gloeocystidia, and chlamydospores; gloeocystidia flexuous, 40–150  $\times$  5–9  $\mu$ , starting from the substratum; imbedded chlamydospores very numerous, globose, 5–6  $\mu$  in diameter, sometimes comprising nearly the whole fructification; basidiospores hyaline, even, 6  $\mu$  in diameter.

Fructifications 3-10 cm. long, 2-4 cm. wide.

On under side of decaying wood and bark of frondose species. Newfoundland and Canada to Louisiana and westward to Washington. September to November. Widely distributed and common locally.

C. effuscatum is conspicuous when fresh by its large salmon to brick-red fructifications. It soon fades in the herbarium to the pallid or buff color assumed in the herbarium by many species and

must then be cautiously separated from C. confluens and Hypochnus pallescens which may have the same aspect. The very numerous imbedded chlamydospores and elongated gloeocystidia of C. effuscatum are its characters for such separation.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 1208.

Newfoundland: Bay of Islands, A. C. Waghorne, 1014 (in Mo. Bot. Gard. Herb., 4805).

Canada: J. Macoun, 16; Lower St. Lawrence Valley, J. Macoun, 3. Ontario: Ottawa, J. Macoun, 455.

Quebec: Ironsides, J. Macoun, 280.

New Hampshire: Chocorua, W. G. Farlow, E (in Mo. Bot. Gard. Herb., 55001).

New York: Ithaca, G. F. Atkinson, 1002, 2113; North Greenbush, H. D. House, 14.236 (in Mo. Bot. Gard. Herb., 44735); Staten Island, W. H. Ballou; Tyre, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57718); Westport, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57770), and 1; White Plains, W. H. Ballou, 1, 2 (in Mo. Bot. Gard. Herb., 55623, 55628).

Pennsylvania: West Chester, Everhart & Haines, in Ellis, N. Am. Fungi, 1208.

District of Columbia: Washington, C. L. Shear, 1262.

Georgia: Tipton, C. J. Humphrey, 162.

Louisiana: A. B. Langlois, 249; St. Martinville, A. B. Langlois, Z. Ohio: C. G. Lloyd, 3824.

Illinois: Bluff Lake, L. H. Pammel (in Mo. Bot. Gard. Herb., 60655).

Missouri: Creve Coeur, E. A. Burt (in Mo. Bot. Gard. Herb., 19458, 44071); Rose Hill, L. H. Pammel (in Mo. Bot. Gard. Herb., 60656); St. Louis, L. H. Pammel, comm. by Farlow Herb.; Upper Creve Coeur, E. A. Burt (in Mo. Bot. Gard. Herb., 54775).

Idaho: Priest River, J. R. Weir, 53.

British Columbia: Vancouver Island, Cedar Hill, J. Macoun.

Washington: Arlington, C. J. Humphrey, 7611 (in Mo. Bot. Gard. Herb., 10750); Kalama, C. J. Humphrey, 6160.

### 59. C. abeuns Burt, n. sp.

Type: in Burt Herb.

Fructification broadly effused, thin, membranaceous, tender, small pieces separable when moistened, whitish to ivory-yellow and cream-buff in the herbarium, even, not cracked or but little cracked, the margin whitish, thinning out, composed of interwoven hyphae; in section  $100-240~\mu$  thick, not colored, composed of somewhat erect, interwoven hyphae  $2\frac{1}{2}-3~\mu$  in diameter, not incrusted, and of slender gloeocystidia; gloeocystidia  $30-60~\times~4-7~\mu$ , numerous, immersed; spores hyaline, even, subglobose,  $6-7~\times~4-6~\mu$ , copious.

Fructifications 4-13 cm. long, 2-5 cm. wide.

On decaying coniferous wood, rarely on bark of frondose species. Maine to Alabama, in British Columbia and New Mexico, and in Japan and South Africa. July to October. Infrequent.

C. abeuns has the aspect of C. lacteum and C. radiosum and spores of nearly the same size as in these species but not quite as globose and further notably distinct from both by its slender, flexuous gloeocystidia.

Specimens examined:

Maine: Piscataquis County, W. A. Murrill, 1938 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 63765).

New Hampshire: North Conway, W. H. Snell, 626 (in Mo. Bot. Gard. Herb., 59293).

New York: Alcove, C. L. Shear, 1215; Freeville, G. F. Atkinson, 2595; Karner, C. H. Peck, comm. by N. Y. State Mus. Herb., T 7 (in Mo. Bot. Gard. Herb., 54557) and another specimen (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55784).

Alabama: Goldbranch, J. R. Weir, 10958 (in Mo. Bot. Gard. Herb., 63240); Montgomery, R. P. Burke, 229, type, and 471 (in Mo. Bot. Gard. Herb., 57100, 57289).

Wisconsin: Madison, M. C. Jensen, comm. by C. J. Humphrey, 617 (in Mo. Bot. Gard. Herb., 44785).

British Columbia: Sidney, *J. Macoun*, 490, 812 (in Mo. Bot. Gard. Herb., 55314, 62117); Squamish, *J. Macoun*, 496 (in Mo. Bot. Gard. Herb., 55184).

New Mexico: Datil National Forest, W. H. Long, 21046 (in Mo. Bot. Gard. Herb., 55145).

Japan: Awaji, A. Yasuda, 12, 80 (in Mo. Bot. Gard. Herb., 55660, 56311).

Africa: Houtbos, Transvaal, P. A. van der Bijl, 1495.

## 60. C. ravum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, closely adnate, thin, not separable, becoming pale pinkish buff to light buff in the herbarium, even, not shining, becoming cracked at the center, the margin thinning out, concolorous; in section 45–150  $\mu$  thick, not colored, composed of densely arranged hyphae, interwoven near substratum but erect towards the hymenium, of numerous gloeocystidia, and of very slender paraphyses; gloeocystidia 20–80  $\times$  7–11  $\mu$ , the more ovoid ones nearer the substratum; paraphyses more or less numerous in the hymenial surface, very slender, hyaline, curved,  $\frac{1}{2}$ -1  $\mu$  in diameter; spores white in spore collection, even, 6-8  $\times$  4–4 $\frac{1}{2}$   $\mu$ .

Fructifications up to 10 cm. long, 2 cm. wide, broken off at both ends.

On fallen frondose limbs. Florida to Louisiana, in Missouri, Cuba, and Brazil. August to February.

C. ravum has grayish fructifications closely resembling well-developed ones of C. rubrocanum in general aspect but distinct by gloeocystidia.

Specimens examined:

Florida: C. G. Lloyd, 4832.

Alabams: Montgomery, R. P. Burke, 126 (in Mo. Bot. Gard. Herb., 5282).

Louisiana: St. Martinville, A. B. Langlois, 1765 and N, type.

Missouri: Creve Coeur, E. A. Burt (in Mo. Bot. Gard. Herb., 44045).

Cuba: Omaja, C. J. Humphrey, 3056.

Brazil: Rio de Janeiro, J. N. Rose, 21462, comm. by N. Y. Bot. Gard. Herb.

## 61. C. mexicanum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb., and N. Y. Bot. Gard. Herb. Fructifications adnate, small, circular, becoming confluent,

rather thick, fleshy-membranaceous, separable when moist, cream color to cream-buff in the herbarium, somewhat velvety or fibrillose, not cracked, the margin concolorous, fimbriate; in section 400  $\mu$  thick, not colored, with hyphae next to the substratum longitudinally and densely arranged, thick-walled, not incrusted, not nodose-septate, curving outward obliquely into the hymenium; gloeocystidia numerous in the hymenium and subhymenium, clavate or cylindric,  $60-120\times 9-12\,\mu$ ; spores few, even, hyaline, not seen attached to basidia,  $9-11\times 6-7\,\mu$ .

Fructifications at first 2-3 mm. in diameter, becoming confluent into a mass 2 cm. long, 5 mm. wide.

On very rotten wood. Mexico. January.

On account of the loose attachment of the fructification to the substratum and the broad layer of longitudinally arranged hyphae it is possible that *C. mexicanum* may be the resupinate stage of a *Stereum*, but if so, it is distinct from any *Stereum* known to me.

Specimens examined:

Mexico: Xuchiles, near Cordoba, W. A. & E. L. Murrill, 1196, type, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54604).

62. C. epigaeum Ell. & Ev. Jour. Myc. 1: 88. 1885; Sacc. Syll. Fung. 6: 631. 1888.

Type: in N. Y. Bot. Gard. Herb.

Fructifications effused, thin, membranaceous, tender, small pieces separable when moistened, white, becoming somewhat pinkish buff in the herbarium, not cracked, the margin concolorous, thinning out; in section 175–250  $\mu$  thick, not colored, 2-layered, the layer next to the substratum about 75  $\mu$  thick, consisting of densely interwoven hyphae about  $2\frac{1}{2}-3$   $\mu$  in diameter, not showing characters clearly in the type; hymenial layer 100–150  $\mu$  thick, composed of densely arranged hyphae, gloeocystidia, and basidia; gloeocystidia elongated; spores hyaline, even,  $5\frac{1}{2}-6\times 5$   $\mu$ , confined to hymenial surface.

Fructifications 2-5 cm. long, 1-3 cm. wide.

On bare ground and rotten wood on the ground. New Hampshire to British Columbia, Washington, and Oregon. August to April. Rare.

 $C.\ epigaeum$  is characterized by white color, 2-layered structure, elongated gloeocystidia, and large, subglobose spores. It is related to  $C.\ lactescens$  but does not become stratose nor cracked nor as hard and compact as the latter. The type specimen itself should be used for comparison rather than the specimens from widely separated localities which seem to me probably to be  $C.\ epigaeum$ .

Specimens examined:

New Hampshire: Chocorua, W. G. Farlow (in Mo. Bot. Gard. Herb., 13954).

New York: Karner, H. D. House, comm. by N. Y. State Mus. Herb., 14.160 (in Mo. Bot. Gard. Herb., 44705).

Ohio: Cincinnati, C. G. Lloyd, 4517.

Michigan: New Richmond, C. H. Kauffman, 20 (in Mo. Bot. Gard. Herb., 9905).

British Columbia: Hastings, J. Macoun, 129. Washington: Bingen, W. N. Suksdorf, 896, 754.

Oregon: Carpenter, 100, type (in N. Y. Bot. Gard. Herb.).

63. C. lactescens Berkeley, Outl. Brit. Fung. 274. 1860; Fries, Hym. Eur. 650. 1874; Sacc. Syll. Fung. 6: 612. 1888; Massee, Linn. Soc. Bot. Jour. 27: 138. 1890; Bresadola, Ann. Myc. 1: 95. 1903; Wakefield, Brit. Myc. Soc. Trans. 4: 118. pl. 3, f. 6-8. 1913; Rea, Brit. Basid. 685. 1922.

Thelephora lactescens Berkeley in Hooker, Eng. Fl. 2: 169. 1836.—Glococystidium lactescens (Berk.) v. Höhnel. & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 784. 1907; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 366. 1913.—Corticium Brinkmanni Bresadola in Brinkmann, Westfälische Prov.-Vereins f. Wiss. u. Kunst Jahresber. 26: 128. 1898.

Type: in Kew Herb.

Fructifications broadly effused, rather thick, closely adnate, waxy-fleshy, small pieces separable, whitish to flesh color and buff-pink when fresh, becoming light buff to avellaneous in the herbarium, even, contracting greatly in drying and forming in thick fructifications very numerous short fissures with somewhat resin-colored sides, the margin whitish, narrow, when fresh exuding a watery white milk where wounded; in section 200–1000 µ

thick, pale avellaneous, becoming stratose when old and thick, with a narrow layer of hyphae arranged longitudinally along the substratum and the remainder of the fructification composed, according to age, of one or more strata of erect, agglutinated hyphae, basidia, and gloeocystidia; gloeocystidia very numerous, flexuous,  $60-120 \times 4-9 \mu$ ; spores hyaline, even, flattened on one side,  $4-8 \times 3-6 \mu$ , copious.

Fructifications 4-10 cm. long, 1-4 cm. wide.

On decaying wood of logs of frondose species. In Europe, Canada to Louisiana, and westward to the Pacific states, in the West Indies, and in Mexico. Throughout the year. Widely distributed but not common.

C. lactescens is best recognized in thick stratose fructifications by their cracking into short and usually disconnected fissures, stratose and agglutinated structure, occurrence on frondose wood, very numerous gloeocystidia, and rather large spores.

Specimens examined:

Exsiccati: Berkeley, Brit. Fungi, 21.

Sweden: Stockholm, L. Romell, 176; Tyresö, L. Romell, C.

Germany: Westfalia, Lengerich, W. Brinkmann, part of type of Corticium Brinkmanni from Bresadola.

Austria: Innsbruck, Tirol, V. Litschauer, 2 specimens; N. Austria, V. Litschauer.

Italy: Trent, G. Bresadola; Pisa, T. Archangeli, comm. by Herb. Horti Pisani (in Mo. Bot. Gard. Herb., 44564).

France: Bois de Boulogne, Paris, G. F. Atkinson.

England: M. J. Berkeley, in Berkeley, Brit. Fungi, 21; West Farleigh, M. J. Berkeley (in Kew Herb.); West Walling, M. J. Berkeley (in Kew Herb.).

Canada: J. Macoun, 12, 20, 81; Hemlock Lake, Beechwood, J. Macoun, 450; Billings Bridge, J. Macoun, 55; Carleton Place, J. Macoun, 91; Lower St. Lawrence Valley, J. Macoun, 26, 32, 36; Ontario, Belleville, J. Macoun, 531.

Newfoundland: Bay of Islands, A. C. Waghorne, 477 (in Mo. Bot. Gard. Herb., 4833).

Maine: Kittery Point, R. Thaxter & E. A. Burt.

Vermont: Middlebury, E. A. Burt; Silver Lake, Leicester, E. A. Burt.

- Massachusetts: Arlington, A. P. D. Piguet, comm. by W. G. Farlow, 34.
- New York: Kirkville, L. M. Underwood, 55 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61566); Ithaca, Thom, comm. by Cornell Univ. Herb., 13725; Vaughns, Hudson Falls, S. H. Burnham, 26 (in Mo. Bot. Gard. Herb., 54492).
- North Carolina: Biltmore Estate, W. A. Murrill (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 61564, and Burt Herb.).
- Louisiana: Baton Rouge, Edgerton & Humphrey, comm. by C. J. Humphrey, 5650.
- Tennessee: Unaka Springs, W. A. Murrill, 623 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61565).
- Michigan: Mass, C. J. Humphrey, 1638 (in Mo. Bot. Gard. Herb., 14228).
- British Columbia: Sidney, J. Macoun, 76, 378, 496 (in Mo. Bot. Gard. Herb., 5752, 55316, 55317).
- Washington: Bingen, W. N. Suksdorf, 909, 911.
- Oregon: Corvallis, S. M. Zeller, 1771, 1905 (in Mo. Bot. Gard. Herb., 56848, 56881).
- California: Pasadena, A. J. McClatchie, 786 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61461).
- Mexico: Jalapa, W. A. & E. L. Murrill, 68, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 1682).
- Porto Rico: Rio Piedras, *J. A. Stevenson*, 3357, 5576 (in Mo. Bot. Gard. Herb., 7688, 11346).
- Grenada: Grand Etang, R. Thaxter, comm. by W. G. Farlow, 16.

# 64. C. salmoneum Burt, n. sp.

Type: in Burt Herb. and in Farlow Herb.

Fructifications broadly effused, adnate, rather thick, somewhat membranaceous, small pieces separable when moist, "orange-salmon" when fresh, becoming vinaceous-buff in the herbarium, even, somewhat velvety, not shining, not at all cracked, the margin similar, determinate, thinning abruptly; in section 360  $\mu$  thick, not colored, composed of densely interwoven hyphae 3–6  $\mu$  in diameter, thin-walled, not incrusted, glued together so that the outline is not clearly defined; gloeocystidia cylindric, up to  $100 \times 8-9 \mu$ , very numerous, confined to the hymenium; spores hyaline, even,  $5 \times 3\frac{1}{2} \mu$ .

Fructifications 4-6 cm. long,  $1\frac{1}{2}-2\frac{1}{2}$  cm. wide, and broken off on three sides in the specimens seen. Probably large.

On bark of decaying frondose wood. West Indies.

This tropical species is somewhat related to *C. lactescens* but differs in not becoming cracked nor stratose and in having its gloeocystidia of nearly equal length and arranged side by side in palisade manner in the hymenial layer.

Specimens examined:

Grenada: Chilly Brook, Grand Etang, R. Thaxter, type, comm. by W. G. Farlow, 16.

## 65. C. Macounii Burt, n. sp.

Type: in Burt Herb.

Fructifications widely effused, closely adnate, soft and fleshy when fresh, drying somewhat cartilaginous, small pieces separable when moistened, white, becoming ivory-yellow in the herbarium, even, sometimes cracking in drying, the margin thinning out; in section  $60\text{--}150~\mu$  thick, not colored, with the hyphae suberect, branching,  $2\frac{1}{2}$ -3  $\mu$  in diameter; gloeocystidia, or perhaps conducting organs, very slender,  $30\text{--}90 \times 3\text{--}3\frac{1}{2}~\mu$ , starting from the substratum; spores hyaline, even, subglobose, slightly flattened on one side,  $8\text{--}10 \times 6\text{--}9~\mu$ , pointed at base, copious.

Fructifications 3-8 cm. long, 1-2 cm. wide.

On decorticated, decaying pine wood. Canada, and perhaps New Hampshire and New York. October. Rare.

C. Macounii is much thinner than C. Berkeleyi and contracts in drying to a horn-like coating on the wood. The gloeocystidia or conducting organs are distinctive but inconspicuous. The specimens from New Hampshire and New York are a little thicker than the Canadian specimens by the presence of a layer of hyphae densely arranged, parallel with the substratum.

Specimens examined:

Canada: Lower St. Lawrence Valley, J. Macoun, 86.

Quebec: Hull, J. Macoun, 368, type.

New Hampshire: Chocorua, W. G. Farlow. New York: Ithaca, G. F. Atkinson, 14102.

# 66. C. argentatum Burt, n. sp.

Type: in Burt Herb.

Fructifications long-effused, thin, closely adnate, not at all separable, pale drab-gray, even, somewhat pruinose, becoming cracked, the margin similar or whitish, thinning out; in section 150  $\mu$  thick, colored buffy brown, composed of densely arranged, interwoven, erect hyphae and gloeocystidia; the hyphae about 3  $\mu$  in diameter, incrusted near the substratum; gloeocystidia very numerous in all regions, usually flexuous,  $40\text{--}50 \times 8\text{--}12 \,\mu$ , but some  $6\text{--}12 \,\mu$  in diameter in the form of spherical brown masses; spores hyaline, even,  $4\text{--}6 \times 3 \,\mu$ —few found and may not belong.

Fructification 10 cm. long, 1 cm. wide.

On under side of small branches of Salix. Nebraska. February. Apparently local.

C. argentatum has aspect so similar to Peniophora cinerea and C. subcinerea that microscopic examination of sections is necessary to separate it from these more common species. Distinguishing characters are the silvery color externally and brown color within and numerous gloeocystidia, some of which have the form of brown spherical masses such as occur in Peniophora serialis.

Specimens examined:

Nebraska: Long Pine, C. L. Shear, 1094, type.

# 67. C. septentrionale Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications broadly effused, adnate, thin, small pieces separable when moist, drying snow-white, waxy, pulverulent, cracking by wide fissures into rectangular masses about  $3\times 2$  mm., the margin similar, composed of interwoven hyphae; in section  $150-200~\mu$  thick, not colored, composed of hyphae loosely arranged below, suberect, bushy-branched, nodose-septate,  $3-3\frac{1}{2}~\mu$  in diameter, not incrusted; gloeocystidia flexuous, up to  $45\times 6~\mu$ , sometimes capitate or moniliform at apex, confined to the hymenial layer; spores hyaline, even, cylindric,  $6-8\times 2-2\frac{1}{2}-3~\mu$ , not numerous; basidia with 4 sterigmata.

Fructifications 5 cm. long, 2 cm. wide, broken off at both ends. On decaying, weathered, frondose wood. Alabama and Manitoba. October.

Among the species having gloeocystidia C. septentrionale is

noteworthy by its snow-white color; the long spores and gloeocystidia with occasionally capitate or moniliform apex may be helpful, confirmatory characters.

Specimens examined:

Alabama: Montgomery Co., R. P. Burke, 672 (in Mo. Bot. Gard. Herb., 63092).

Manitoba: Winnipeg, G. R. Bisby, 1346, type (in Mo. Bot. Gard. Herb., 60556).

68. C. stramineum Bresadola, Hedwigia 39: (221). 1900; Sacc. Syll. Fung. 16: 193. 1902.

Gloeocystidium stramineum Bresadola in Brinkmann, Westfälische Pilze, 18; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 361. 1913.—See Wakefield, Brit. Myc. Soc. Trans. 4: 341. 1918.

Type: type distribution in Brinkmann, Westfälische Pilze, 18. Fructifications broadly effused, adnate, thin, somewhat membranaceous, small pieces separable when moist, becoming cartridge-buff to cream-buff in the herbarium, even, becoming somewhat cracked, the margin thinning out, pruinose, similar; in section  $100-200\,\mu$  thick, not colored, composed of suberect, interwoven hyaline hyphae 2-3  $\mu$  in diameter, not incrusted, and of elongated gloeocystidia; gloeocystidia flexuous, tapering towards apex,  $40-100\times41/2-9\,\mu$ ; spores hyaline, even,  $4-6\times2-3\,\mu$ , not copious.

Fructifications 2-8 cm. long, 1-3 cm. wide.

On bark of decaying Alnus, Acer rubrum, and Carya. In Europe, and from Canada to South Carolina and westward to British Columbia and in Mexico. September to January. Rare.

C. stramineum may be recognized among our species having gloeocystidia, by its thin, whitish to straw-colored fructification on Acer rubrum.

Specimens examined:

Sweden: L. Romell, 419.

Germany: Lengerich, Westphalia, W. Brinkmann, part of type from Bresadola.

Austria: Tirol, V. Litschauer, 4 specimens from Innsbruck, Klosterberg, Stubai, and Volders, respectively.

Canada: J. Macoun, 28; Ontario, Ottawa, J. Macoun, 18.

New Hampshire: Chocorua, W. G. Farlow, 31 and unnumbered specimen.

Vermont: Middlebury, E. A. Burt.

New York: Bronx Park, L. M. Underwood (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61594); Ithaca, G. F. Atkinson, 3087; Karner, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 54365, 54362).

New Jersey: Newfield, J. B. Ellis, comm. by N. Y. Bot. Gard. Herb.

Maryland: Takoma Park, C. L. Shear, 1338.

South Carolina: Hartsville, W. C. Coker, 3947 (in Mo. Bot. Gard. Herb., 57415).

Keniucky: Crittenden, C. G. Lloyd, 3124.

Missouri: Creve Coeur, F. P. McWhorter (in Mo. Bot. Gard. Herb., 57451).

British Columbia: Sidney, J. Macoun, 74, 80, in part (in Mo. Bot. Gard. Herb., 5749, 5750).

Mexico: Orizaba, W. A. & E. L. Murrill, 763, comm. by N. Y. Bot. Gard. Herb., 54634.

# 69. C. Litschaueri Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, adnate, thin, somewhat membranaceous, small pieces separable when moistened, between ivory-yellow and olive-buff in the herbarium, even, becoming somewhat cracked, not shining, the margin thinning out; in section 200  $\mu$  thick, not colored, composed of loosely interwoven, thick-walled hyphae 3  $\mu$  in diameter, nodose-septate, not incrusted; gloeocystidia flexuous,  $45-120 \times 4\frac{1}{2}-6 \mu$ , in all regions of the fructification; spores hyaline, even, cylindric, flattened on one side,  $9-10 \times 3-3\frac{1}{2} \mu$ , four to a basidium.

Fragments of fructification 2 cm. long,  $1-1\frac{1}{2}$  cm. wide, broken off on three sides.

On bark of Alnus and apple. North Dakota and Oregon.

C. Litschaueri has the aspect of C. stramineum and occurs on a frequent substratum of the latter but the spores of C. Litschaueri are the larger and the hyphae are thicker-walled than those of C. stramineum and more like those of P. cremea.

Specimens examined:

North Dakota: Brenckle, comm. by V. Litschauer, 1, type.

Oregon: Corvallis, S. M. Zeller, 2219 (in Mo. Bot. Gard. Herb., 63029).

## 70. C. protrusum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications broadly effused, rather thick, dry, felty-membranaceous, separable, drying between light buff and cream color, even, conforming to irregularities of the substratum, not cracked, the margin a little paler than the hymenium, thinning out, with the hyphae interwoven; in section 500  $\mu$  thick, not colored, 2-layered, with (1) a broad layer next to the substratum of very densely and longitudinally arranged hyphae, and with (2) a somewhat more loosely arranged layer of interwoven, suberect, hyaline hyphae  $4-4\frac{1}{2}\mu$  in diameter, not incrusted, and occasional gloeocystidia; gloeocystidia flexuous, up to  $60 \times 4\frac{1}{2}-5\mu$ ; basidia 4-spored, not side by side and adhering together in a compact palisade layer but very numerous and protruding individually  $6-15\mu$ ; spores attached to basidia are hyaline, even,  $6 \times 2\frac{1}{2}-3\mu$ , tapering towards the base, not copious.

Fructification 6 cm. long, 5 cm. wide, broken off on one side and at one end—probably large.

On bark of a badly decayed frondose log in a moist virgin forest. Mexico. December.

C. protrusum has a large fructification of general aspect and color of that of C. portentosum and C. galactinum but softer than these, of quite different structure, and having gloeocystidia. The basidia protrude beyond the general level of the fructification in the manner of cystidia; the presence of spores at the apex shows convincingly their real nature.

Specimens examined:

Mexico: Jalapa, W. A. & E. L. Murrill, 157, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 10354).

71. C. livido-caeruleum Karsten, Notiz ur Sällsk. pro Faun. et Fl. Fenn. Förh. 9: 370. 1868; Finska Vet.-Soc. Bidrag Natur och Folk 25: 315. 1876; 48: 415. 1889; Icones Hym. Fenniae

3: 8. f. 75. 1889; Fries, Hym. Eur. 652. 1874; Sacc. Syll. Fung. 6: 623. 1888; Massee, Linn. Soc. Bot. Jour. 27: 152. 1890.

Gloeocystidium livido-caeruleum (Karst.) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1554. 1906; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 355. 1913.—An Corticium plumbeum Fries, Hym. Eur. 653. 1874?

Type: studied from Karsten Herb. in Helsingfors by v. Höhnel & Litschauer, loc. cit.

Fructifications long-effused, agglutinate, waxy-soft, not separable, white at first, then darkening in spots, finally more or less completely slate-gray to dark plumbeous, white, pruinose, rarely cracked; in section 100–250  $\mu$  thick, colored within when mature by 1–3 bluish black layers whose color is unchanged in lactic acid mounts but becomes at first vinaceous and is then dissolved and the sections bleached by potassium hydrate solution; very young fructifications not colored within; hyphae about 3  $\mu$  in diameter, glued together so that the outline is not clearly shown; gloeocystidia elongated, flexuous, 30–60  $\times$  3–6  $\mu$ ; spores hyaline, even,  $4\frac{1}{2}$ –6  $\times$   $2\frac{1}{2}$ – $3\frac{1}{2}$   $\mu$ .

Fructifications 1-10 cm. long, 2 mm.-3 cm. wide.

Under side of decaying coniferous rails, boards, and shingles, recorded on *Abies*, *Pinus* and *Thuja*. In Europe and in Canada, Vermont, New York, Montana, and Manitoba. April to September. Infrequent.

The dark lead color of one or more layers in the interior of the fructifications and the destruction of the coloring pigment by seven per cent potassium hydrate solution, together with the presence of gloeocystidia, afford a group of characters by which C. livido-caeruleum may be confidently recognized. Karsten did not send me an authentic specimen of his C. livido-caeruleum but he sent a specimen with the same characters under the name Corticium plumbeum Fr.

Specimens examined:

Sweden: L. Romell, 107; Lappland, L. Romell, 409.

Finland: Mustiala, P. A. Karsten, under the name C. plumbeum Fr.

Austria: Tirol, Innsbruck, V. Litschauer; Stubai, V. Litschauer. Canada: J. Macoun, 37.

Vermont: Middlebury, E. A. Burt, 2 gatherings.

New York: Altamont, E. A. Burt.

Montana: Fontine, E. E. Hubert, comm. by J. R. Weir (in Mo. Bot. Gard. Herb., 63234); Missoula, J. R. Weir, 420 (in Mo. Bot. Gard. Herb., 14767), and E. E. Hubert, comm. by J. R. Weir, 11961 (in Mo. Bot. Gard. Herb., 63318); Trego, E. E. Hubert, comm. by J. R. Weir, 11975 (in Mo. Bot. Gard. Herb., 63331).

Idaho: Avery, E. E. Hubert, comm. by J. R. Weir, 11987 (in Mo. Bot. Gard. Herb., 63320).

Manitoba: Norway House, G. R. Bisby, 1462 (in Mo. Bot. Gard. Herb., 61644).

British Columbia: Kootenai Mts., near Salmo, J. R. Weir, 466 (in Mo. Bot. Gard. Herb., 14936).

#### 72. C. pilosum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, becoming confluent, closely adnate, very thin, not separable, pale pinkish buff, becoming pale olivebuff and pale smoke-gray in the herbarium, even, not shining, but little or not at all cracked, the margin of the same color, thinning out; in section  $30-75\,\mu$  thick, not colored, composed of densely interwoven, hyaline hyphae  $2-2\frac{1}{2}\,\mu$  in diameter, not incrusted, of gloeocystidia, and of delicate, branching paraphyses; gloeocystidia near the substratum, spherical or pyriform,  $16-30\,\mu$  in diameter or up to  $30\,\times\,15\,\mu$ , narrower gloeocystidia may be present also; paraphyses with slender branching tips about  $1\,\mu$  in diameter occur in the surface of the hymenium; spores hyaline, even, curved,  $6-9\,\times\,3-4\frac{1}{2}\,\mu$ .

Fructifications becoming confluent over areas up to 8 cm. long and 1-2 cm. wide.

On bark of fallen limbs of *Alnus*, *Vitis*, and *Tsuga*. Georgia, Alabama and Missouri. October and April. Not common.

C. pilosum has general aspect and color suggestive of the Peniophora cinerea group of species but has no cystidia. The slender branching paraphyses have been noted also in Peniophora phyllophila, C. albido-carneum, C. Atkinsonii, and C. jamaicense. Perhaps C. pilosum is mature C. albido-carneum.

Specimens examined:

Georgia: Atlanta, E. Bartholomew, 8982, type (in Mo. Bot. Gard. Herb., 63463).

Alabama: Auburn, Earle & Baker (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 63709, 61479); Montgomery, R. P. Burke, 16, 217, 350, 452, 613 (in Mo. Bot. Gard. Herb., 4738, 57089, 57221, 57275, 57443).

Missouri: Baden, E. A. Burt (in Mo. Bot. Gard. Herb., 18864).

73. C. radiosum Fries, Epicr. 560. 1838; Icones Hym. 2: 97. pl. 198, f. 1. 1884; Hym. Eur. 649. 1874; Sacc. Syll. Fung. 6: 611. 1888; Bresadola, I. R. Accad. Agiati Atti III. 3: 110. 1897; Rea, Brit. Basid. 685. 1922.

Thelephora radiosa Fries, Obs. Myc. 2: 277. 1818; Elench. Fung. 1: 206. 1828; Persoon, Myc. Eur. 1: 130. 1822.—
Corticium pellicula (Fr.) Karsten, Soc. pro Fauna et Fl. Fenn. Meddel. 11: 5. 1885.—Corticium alutaceum (Schrad.) Bresadola, I. R. Accad. Agiati Atti III. 3: 110. 1897; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 115: 1556. 1906.

—Gloeocystidium alutaceum (Schrad.) Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 367. 1913.—An Thelephora alutacea Schrader, Spic. Fl. Germ. 1: 187. 1794?

Type: type illustration is Fries, Icones Hym. 2: pl. 198, f. 1. 1884. No authentic specimen determined, by E. Fries as *Thelephora* (or *Corticium*) radiosa is known.

Fructifications broadly effused, thin, membranaceous, tender, small pieces separable, from whitish to ivory-yellow and creambuff in the herbarium, even, but little cracked, the margin white, broad, radiating, fibrillose; in section 100–300  $\mu$  thick, not colored, composed of densely interwoven, ascending hyphae rather crowded together except where separated by vesicular bodies which become greatly inflated and thin-walled and are finally up to  $20-60 \times 15 \,\mu$ ; spores hyaline, even or slightly rough, subglobose,  $4\frac{1}{2}-7 \,\mu$  in diameter or  $6 \times 4\frac{1}{2}-5 \,\mu$ .

Fructifications 3-15 cm. long, 1-7 cm. wide.

On decaying wood of coniferous species usually. In Europe, Canada to Pennsylvania, and westward to Alaska, British Columbia, and Washington.

 $C.\ radiosum$  may be recognized by its occurrence on coniferous wood, whitish or ivory-yellow color, white fimbriate margin, subglobose spores about  $6~\mu$  in diameter, and presence of very large vesicular bodies when sections are examined. These bodies are often so inflated and with walls so tenuous that their location is shown by vesicular spaces between the otherwise crowded hyphae.

No authentic specimen of C. radiosum determined by E. Fries is known to be in existence, although there are four specimens so determined by Karsten in Herb. Fries; two of these specimens are Peniophora laevis, another is very immature but may be Stereum odoratum, while the fourth specimen, Karsten, No. 32, has globose spores  $6-8 \times 5-6 \mu$  but does not show vesicular bodies in my mount. However, these four specimens present the Karsten idea of C. radiosum as to aspect. The colored illustration of C. radiosum in Fries' Icones, pl. 198, f. 1, is excellent, and taken in connection with the good original description by Fries and his critical comment on the close resemblance to his Peniophora laevis. seems to me to afford a more secure foundation for the concept of this species as C. radiosum than as Corticium alutaceum, for Schrader's description of Thelephora alutacea consists of the following five words, viz., "Supra exalbida, subtus tomentosa nivea." This vague description is not supplemented by an illustration, and I have not been able to learn of the existence of an authentic specimen. Any statement as to synonymy in the case of resupinate Hymenomycetes by mycologists of a former century is of slight value when a nice feature of internal structure is decisive.

Specimens examined:

Exsiccati: Ell. & Ev., Fungi Col., 1211, under the name Corticium Petersii.

Sweden: Femsjö, L. Romell, 177; Stockholm, L. Romell, 113, 178, 181.

Austria: Innsbruck, Tirol, V. Litschauer; Stubai, Tirol, V. Litschauer, 2 specimens—all as C. alutaceum.

Hungary: Tatra Magna, V. Greschik, from Bresadola, under the name C. alutaceum.

Canada: Lower St. Lawrence Valley, J. Macoun, 87; Ontario, Ottawa, J. Macoun, 133, 204.

- Vermont: Bethel, P. Spaulding, comm. by U. S. Path. & Myc. Coll., 2708; Middlebury, E. A. Burt, 2 gatherings.
- Massachusetts: Sharon, A. P. D. Piguet (in Farlow Herb., 127, and Mo. Bot. Gard. Herb., 55234).
- New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 59672); Constableville, C. H. Peck, comm. by N. Y. State Mus. Herb., T 3 (in Mo. Bot. Gard. Herb., 54556, 55774); Fort Ann, S. H. Burnham, 11 (in Mo. Bot. Gard. Herb., 54508); Freeville, G. F. Atkinson, 2585; Ithaca, G. F. Atkinson, 2527, 14186; Schuylerville, C. H. Peck, 19, and an unnumbered specimen (in N. Y. State Mus. Herb., 55772).
- New Jersey: Newfield, J. B. Ellis (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 54450).
- Pennsylvania: Bellefonte, L. O. Overholts, 3729 (in Mo. Bot. Gard. Herb., 55098); State College, C. R. Orton, 2, comm. by L. O. Overholts (in Mo. Bot. Gard. Herb., 44041).
- West Virginia: Nuttallburg, L. W. Nuttall, in Ell. & Ev., Fungi Col., 1211.
- Tennessee: Elkmont, C. H. Kauffman, 89 (in Mo. Bot. Gard. Herb., 44990).
- Michigan: Ann Arbor, C. H. Kauffman, 36 (in Mo. Bot. Gard. Herb., 19327); East Lansing, E. A. Bessey (in Mo. Bot. Gard. Herb., 56178); New Richmond, C. H. Kauffman, 50 (in Mo. Bot. Gard. Herb., 18523).
- Missouri: Creve Coeur L. O. Overholts (in Mo. Bot. Gard. Herb., 42602).
- Arkansas: Fordyce, C. J. Humphrey, 2528 (in Mo. Bot. Gard. Herb., 14057).
- Washington: Bellingham, J. R. Weir, 546 (in Mo. Bot. Gard. Herb., 63744); Olympic Mts., comm. by W. G. Farlow, 3 (in Mo. Bot. Gard. Herb., 44588); Sedro-Woolley, C. J. Humphrey, 7483.
- British Columbia: Sidney, J. Macoun, 25 (in Mo. Bot. Gard. Herb., 5686).
- Alaska: Ketchikan, J. R. Weir, 329 (in Mo. Bot. Gard. Herb., 16437).

## 74. C. vesiculosum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, closely adnate, thin, between ivory-yellow and cream color in the herbarium, waxy, even, not cracked, the margin thinning out; in section 150–240  $\mu$  thick, not colored, somewhat stratose, with the 3 strata or layers of the type separated by narrow zones of hyphae glued together; hyphae about 2  $\mu$  in diameter, thin-walled, collapsing, poorly defined, densely interwoven; gloeocystidia up to 40  $\times$  8  $\mu$ ; many vesicular bodies, presumably gloeocystidia, are present and are 5–7  $\mu$  in diameter—also larger vesicular spaces; spores hyaline, even,  $4-8\times2^{1}/_{2}-4$   $\mu$ , borne on protruding basidia having 4 sterigmata.

Fructifications in fragments up to 4 cm. long,  $1\frac{1}{2}$  cm. wide.

On decaying, frondose wood. Canada and New York. October.

C. vesiculosum is colored like C. radiosum but is closely adnate, does not have a radiating, fibrillose margin, and has smaller spores.

Specimens examined:

Canada: J. Macoun, 71, type.

New York: East Galway, E. A. Burt.

# 75. C. globosum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, thick, adnate, spongy-soft, white, becoming cartridge-buff, somewhat granular, not waxy, cracked only rarely, the margin determinate, thick, with hyphae interwoven; in section 600–700  $\mu$  thick, grayish olive near the substratum, stratose, each stratum composed of slightly colored, thinwalled, suberect, curving and branching hyphae  $\frac{1}{2}-1$   $\mu$  in diameter, and of scattered, conspicuous, rather thick-walled, globose vesicular bodies 12–13  $\mu$  in diameter; no other gloeocystidia; no cystidia; spores hyaline, even,  $3 \times 2 \mu$ .

Largest fragments of fructifications are 3 cm. in diameter and 4 cm. long, 2 cm. wide.

On rotten frondose wood. West Indies. November. Probably local.

C. globosum forms thick, pulvinate fructifications suggestive in

aspect of those of resupinate Stereum Murrayi but soft and spongy when moistened. The abundant, slender, curving hyphae show structural relationship with Corticium investiens and Hyphochnus pallescens, but I find no antler-shaped branches either at the hymenial surface or in the interior. The globose vesicular bodies are conspicuous and a valuable distinctive character.

Specimens examined:

Cuba: Omaja, C. J. Humphrey, 2842.

Porto Rico: Rio Piedras, J. A. Stevenson, 5793, type (in Mo. Bot. Gard. Herb., 54690), and J. A. Stevenson & R. C. Rose, 6531 (in Mo. Bot. Gard. Herb., 55652).

## 76. C. subalbum Burt, n. sp.

Type: in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., an Burt Herb.

Fructifications effused, very thin, closely adnate, whitish to cartridge-buff in the herbarium, even, not shining, but little cracked, the margin similar, thinning out; in section 75  $\mu$  thick, not colored, composed of densely interwoven hyphae about 2  $\mu$  in diameter, and of very numerous gloeocystidia which are broadly ovoid to subglobose, up to 30  $\times$  15–18  $\mu$ , or 20  $\mu$  in diameter; very slender paraphyses with branched tips protrude slightly beyond the basidia; spores hyaline, even,  $10\frac{1}{2}$ –13  $\times$  4–5  $\mu$ , copious.

Fructifications 3-5 mm. in diameter, clustered near together and becoming confluent in a mass 5 cm. long, 1½ cm. wide.

On small dead limbs of *Alnus*. Georgia and Alabama. November.

C. subalbum is distinct from other gloeocystidial species by thin, whitish fructifications, rather large spores, abundant gloeocystidia, and the slender paraphyses.

Specimens examined:

Georgia: Atlanta, E. Bartholomew, 8983 (in Mo. Bot. Gard. Herb., 63462).

Alabama: Auburn, F. S. Earle, 2300, type (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 63375, and Burt Herb.).

# 77. C. vinososcabens Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, adnate, rather thick, membranaceous, separable when moistened, vinaceous-buff or pale avellaneous when fresh, becoming deep purplish vinaceous where bruised, finally between pale olive-buff and pale pinkish buff in the herbarium, even, waxy, not cracking, the margin whitish, fimbriate; in section 150–450  $\mu$  thick, pale-colored, with a compact hymenial layer containing numerous thin-walled, vesicular bodies 15–75  $\times$  12–45  $\mu$ , and with a very broad supporting layer consisting of thin-walled, nodose-septate hyphae 2–3  $\mu$  in diameter, not incrusted and loosely arranged except in thick fructifications where 1 or 2 dense narrow zones are present between substratum and hymenial layer; basidia 2-spored; spores white in spore collection, even, subglobose, 6–9  $\times$  5–7  $\mu$ , slightly pointed at the base.

Fructifications 3-9 cm. long, 1½-3 cm. wide.

On bark of fallen trunk of *Abies rubra* and *Tsuga canadensis*. Vermont to Wisconsin. September and November. Rare.

C. vinososcabens dries a characteristic livid color, occurs on bark of conifers, and has large subglobose spores and a vesiculose hymenial layer. These vesicular organs are presumably gloeocystidia but so highly inflated that they appear empty under the microscope, and with their scanty cell contents adhering to the cell wall.

Specimens examined:

Vermont: Little Notch, Ripton, E. A. Burt, type.

New York, Karner, H. D. House, 14.210 (in Mo. Bot. Gard. Herb., 44730).

Wisconsin: Ladysmith, C. J. Humphrey, 1773 (in Mo. Bot. Gard. Herb., 14242).

78. C. polygonium Persoon, Roemer Neues Mag. Bot. 1: 110. 1794; Fries, Epicr. 564. 1838; Hym. Eur. 655. 1874; Berkeley, Outl. Brit. Fung. 276. 1860; Sacc. Syll. Fung. 6: 627. 1888; Massee, Linn. Soc. Bot. Jour. 27: 144. 1890; Bresadola, Ann. Myc. 1: 97. 1903; Rea, Brit. Basid. 684. 1922.

Thelephora polygonia Persoon, Syn. Fung. 574. 1801; Myc. Eur. 1: 132. 1822; Fries, Syst. Myc. 1: 444. 1821; Elench. Fung. 1: 222. 1828.—Gloeocystidium polygonium (Pers.) v. Höhnel & Litschauer, Wiesner Festschr. Wien, 69. 1908;

Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 363. 1913.—G. polygonium (Pers.) var. fulvescens Bresadola, Mycologia 17: 69. 1925.

Fructifications orbicular, soon confluent and broadly effused, closely adnate, thin, pale ecru-drab to brownish drab, pruinose, even or somewhat tubercular, waxy, the margin whitish; in section 150-250  $\mu$  thick, not colored, composed of suberect, interwoven hyphae 3-5  $\mu$  in diameter, occasionally nodose-septate, and of pyriform gloeocystidia  $10-25 \times 5-20 \mu$ ; spores hyaline, even, cylindric, slightly curved,  $7\frac{1}{2}-10 \times 2\frac{1}{2}-3 \mu$ .

Fructifications 3-5 mm. in diameter, becoming by confluence up to 8 cm. long, 1-2 cm. wide.

On fallen branches of *Populus*. In Europe and in Colorado, Idaho, Manitoba, and Washington.

American specimens of *C. polygonium* are not as heavily pruinose as the European specimens which I have seen and may be recognized by the light grayish vinaceous color of the fructifications, occurrence on poplar bark, large, scattered gloeocystidia, and slender, cylindric spores.

Specimens examined:

Exsiccati: Cooke, Fungi Brit., 6; Romell, Fungi Scand., 128.

Sweden: Stockholm, L. Romell, 118, 119, and in Romell, Fungi Scand., 128, and W. A. Murrill, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 61477); Svex, Söderm., Lindblad, from E. Fries (in Kew Herb.).

Germany: Brinkmann, comm. by Bresadola.

Austria: Tirol, V. Litschauer.

England: Batheaston, C. E. B., in Cooke, Fungi Brit., 6.

Colorado: Geneva Creek Canyon, F. J. Seaver & E. Bethel (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61474); Lake Eldora, F. J. Seaver & E. Bethel (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 56793).

Idaho: J. R. Weir, 16824, type of Gloeocystidium polygonium var. fulvescens (in Weir Herb.); Coolin, J. R. Weir, 11551 (in Mo. Bot. Gard. Herb., 63703); Priest River, J. R. Weir, 14946 (in Mo. Bot. Gard. Herb., 56803).

Manitoba: I. L. Conners & J. F. Higham, comm. by G. R. Bisby, 394 (in Mo. Bot. Gard. Herb., 58969).

Washington: Bingen, W. N. Suksdorf, 719, 902.

79. C. chrysocreas Berk. & Curtis, Grevillea 1: 178. 1873; Sacc. Syll. Fung. 6: 618. 1888.

Corticium crocicreas Massee, Linn. Soc. Bot. Jour. 27: 151. 1890; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 776. 1907.—Not C. crocicreas Berk. & Curtis.

Type: type distribution in Ravenel, Fungi Car. 5:27, under the name Corticium crocicreas.

Fructifications broadly effused, rather thick, closely adnate, not at all separable, apricot-yellow and olive-ocher to dark olive-buff, even or becoming somewhat papillate, cracked in drying, the margin thinning out, indeterminate; in section 120–300  $\mu$  thick, olive-ocher throughout, composed of erect, densely interwoven and conglutinate colored hyphae about 2  $\mu$  in diameter, of very numerous vesicular organs 15–21  $\times$  6–9  $\mu$ ; coloring matter of the sections becomes vinaceous upon treatment with potassium hydrate solution and the sections are finally bleached; spores white in a spore collection, even,  $4\frac{1}{2}$ –5  $\times$   $2\frac{1}{2}$   $\mu$ .

Fructifications 3-8 cm. long, 1-3 cm. wide.

On wood and bark of decaying logs of frondose species. South Carolina to Louisiana and Missouri, in Mexico, in West Indies, and in Japan. July to April. Occasional.

C. chrysocreas has olive-ocher fructifications of the same color throughout which make it one of the most conspicuous species of the region bordering on the Gulf of Mexico. Several other Gulf species have a northern station in Missouri or Illinois. The vesicular structure in section is an important distinctive character for separation of this species from Odontia Wrightii, which has the same color and geographical range but angular granules in the hymenium.

Specimens examined:

Exsiccati: Ell. & Ev., N. Am. Fungi, 2021, under the name Corticium crocicreas—in some copies this, and in others a different species; Ravenel, Fungi Car. 5:27, under the name C. crocicreas.

South Carolina: H. W. Ravenel, Curtis Herb., 2933, type (in Kew Herb.) and in Ravenel, Fungi Car. 5: 27.

Florida: W. W. Calkins, in some copies of Ell. & Ev., N. Am. Fungi, 2021; New Smyrna, C. G. Lloyd, 2072.

Alabama: Peters, 418 (under the name C. crocicreas in Curtis Herb., 4027).

Mississippi: Hattiesburg, C. J. Humphrey, 5454.

Louisiana: Baton Rouge, Edgerton & Humphrey, comm. by C. J. Humphrey, 5601; St. Martinville, A. B. Langlois, bm, H. 2612, and 35—the last comm. by Lloyd Herb., 2386—and 1950a, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 42601).

Missouri: Creve Coeur, E. A. Burt (in Mo. Bot. Gard. Herb., 1757, 14199).

Mexico: Jalapa, W. A. & E. L. Murrill, 180, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 44968).

Cuba: Baracoa, L. M. Underwood & F. S. Earle, 1210, comm. by N. Y. Bot. Gard. Herb.

Japan: Hida-Machi, Prov. Bungo, N. Nakayama, comm. by A. Yasuda, 96, under the name Corticium Nakayamae Yasuda.

## 80. C. involucrum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, closely adnate, thin, somewhat gelatinous, not at all separable, drying olive-buff to snuff-brown, even, conforming to inequalities of the substratum, pruinose, not cracked except where bridging a depression, the margin indeterminate, thinning out; in section 60–80  $\mu$  thick when composed of 1 stratum, 120–150  $\mu$  when 2 strata are present, colored like the hymenium by the color of the numerous gloeocystidia, each stratum composed of erect, densely arranged hyphae and gloeocystidia; hyphae 3  $\mu$  in diameter, with outer wall somewhat gelatinously modified, clothed with short lateral branches up to 6  $\mu$  long which are clustered in an involucral cup at the base of the basidium; gloeocystidia brownish-colored, irregular, flexuous, 30–45  $\times$  4–4½  $\mu$ , very numerous; basidia simple, bearing 4 spores; spores hyaline, even, spherical, 3–4  $\mu$  in diameter.

Fructifications 2-10 cm. long, 1-3 cm. wide.

Under side of decorticated, decaying logs of frondose species usually—one gathering on coniferous wood. Canada, New Hampshire, Vermont, and Cuba. September to December.

C. involucrum forms a thin brown coating on decaying wood, with aspect somewhat suggestive of a Sebacina or C. lividum but so near the color of the wood and so inconspicuous that it is probably often overlooked; the colored gloeocystidia are addi-

tional confirmatory characters which should identify the species. The hyphal structure is unique but not likely to be observed unless close study is made.

Specimens examined:

Canada: Ottawa, J. Macoun, 4, 23.

New Hampshire: Chocorua, W. G. Farlow, 7.

Vermont: Middlebury, E. A. Burt, type.

Cuba: Ceballos, C. J. Humphrey, 2793 (in Mo. Bot. Gard. Herb., 20200).

81. C. luridum Bresadola, Fungi Trid. 2:59. pl. 169. 1898; Sacc. Syll. Fung. 16:119. 1902.

Gloeocystidium luridum (Bres.) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 770. 1907; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 360. 1913.

Type: part of type in Burt Herb.

Fructifications broadly effused, adnate, sometimes rather thick, small pieces separable when moistened, becoming cinnamon-buff to avellaneous in the herbarium, not shining, even, sometimes somewhat cracked with age, the margin similar; in section 150–300  $\mu$  thick, slightly colored, composed of densely arranged hyphae 2–3½  $\mu$  in diameter and not incrusted, which run parallel with the substratum in a narrow layer and then become erect and mixed with gloeocystidia in a broad layer which bears the hymenium; gloeocystidia numerous, slightly colored, flexuous, 50–100  $\times$  6–7  $\mu$ ; spores hyaline, even, 6–8  $\times$  3–5  $\mu$ .

Fructifications 3-4 cm. long, 1-4 cm. wide and broken off at both ends in the fragments received.

On bark and wood of frondose species. In Europe, Ohio, and Manitoba. Autumn. Rare.

C. luridum may be recognized among our species by its slightly colored gloeocystidia and resemblance in general aspect and color to Peniophora velutina. The spores were published by Bresadola as  $10\text{--}17 \times 6\text{--}8~\mu$  but I have found none so large in the specimen received.

Specimens examined:

Italy: Florentia, Martelli, type, from Bresadola.

Ohio: Preston, C. G. Lloyd, 1558.

Manitoba: Winnipeg: A. H. R. Buller, 744 (in Mo. Bot. Gard. Herb., 57913).

#### 82. C. jamaicense Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, adnate, thick, somewhat membranaceous, small pieces separable when moistened, becoming buff-brown to tawny olive in the herbarium, even, pulverulent, not cracked, the margin probably thick and entire but not well shown by the fragments; in section 150–600  $\mu$  thick, concolorous with the hymenium, composed of even, suberect hyphae 3  $\mu$  in diameter, of interwoven organs 2  $\mu$  in diameter with antler-shaped branching, of colored gloeocystidia, and of imbedded, globose, slightly colored, rough-walled spores 6–7  $\mu$  in diameter, very numerous in all regions; gloeocystidia becoming dark-colored, irregular, flexuous, 35–60  $\times$  5–7  $\mu$ , scattered throughout the fructifications, none found protruding; basidia simple, with 4 sterigmata; basidiospores spherical, hyaline, even, 6  $\mu$  in diameter as seen attached to basidia.

Fructifications received in fragments, of which the largest is 7 cm. long, 2 cm. wide.

On decaying wood. Jamaica. December to January.

The general aspect and antler-shaped branching of one kind of its hyphal components show relationship to *Hypochnus penio-phoroides*, *H. pallescens*, *Stereum induratum*, *S. duriusculum*, *Asterostromella dura*, and *A. rhodospora*. Could I have found uneven basidiospores this species would have been included in *Hypochnus* near *H. pallescens* and *H. peniophoroides*; such basidiospores may eventually be demonstrated when this species becomes better known.

Specimens examined:

Jamaica: Cinchona, W. A. & E. L. Murrill, 456, type, comm. by N. Y. Bot. Gard. Herb.; Morce's Gap, W. A. & E. L. Murrill, 677, 740, comm. by N. Y. Bot. Gard. Herb.

83. C. debile Berk. & Curtis in Massee, Linn. Soc. Bot. Jour. 27: 131. 1890; Sacc. Syll. Fung. 11: 127. 1895.

Type: in Kew Herb. and Farlow Herb.

Fructifications broadly effused, thin, closely adnate, becoming pale ivory-yellow to buffy brown in the herbarium, even, waxy, not cracked, the margin whitish; in section 150  $\mu$  thick, yellowish by presence of numerous colored gloeocystidia, with the hyphae about  $2\frac{1}{2}-3$   $\mu$  in diameter, with walls gelatinously modified and poorly defined, longitudinally arranged along substratum and then ascending to the hymenium; gloeocystidia somewhat colored, flexuous,  $30-60 \times 3-5$   $\mu$ ; some colorless vesicular bodies present also; spores hyaline, even, subglobose, 4-5  $\mu$  in diameter in Burt preparation but noted by Massee as  $7 \times 3-4$   $\mu$ .

Fructifications 1-3 cm. in diameter.

Under side of decaying frondose limbs on the ground. Louisiana, California, West Indies, and Venezuela. June and December. Rare.

C. debile has gloeocystidia which are numerous and conspicuous by their yellowish color; these gloeocystidia and the brown fructifications afford good distinguishing characters.

Specimens examined:

Louisiana: St. Martinville, A. B. Langlois, bb, 2674 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 58327).

California: Preston's Ravine, Palo Alto, W. A. Murrill & L. S. Abrams, 1195, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 55709).

Cuba: near Havana, C. J. Humphrey, 2963.

Porto Rico: Bayamon, J. A. Stevenson, 6762 (in Mo. Bot. Gard. Herb., 55053); Rio Piedras, J. A. Stevenson, 5620, and J. A. Stevenson & R. C. Rose, 6529 (in Mo. Bot. Gard. Herb., 44864, 55082); Martin Peña, J. A. Stevenson, 3719 (in Mo. Bot. Gard. Herb., 7091).

Jamaica: Constant Spring Hotel grounds, W. A. & E. L. Murrill, 26, comm. by N. Y. Bot. Gard. Herb.

Venezuela: Fendler, type (in Curtis Herb., 204).

84. C. venosum Berk. & Ravenel, Grevillea 1: 177. 1873; Sacc. Syll. Fung. 6: 620. 1888; Massee, Linn. Soc. Bot. Jour. 27: 147. 1890.

Type: in Kew Herb. and Farlow Herb.

Fructifications broadly effused, rather thick, waxy-gelatinous

when moistened, becoming vinaceous-brown in the herbarium, even; in section 500–600  $\mu$  thick, with a layer 300  $\mu$  broad towards the substratum composed of longitudinally and densely arranged hyphae, with the outer walls so gelatinously modified that only the stained lumen and contents of each hypha are now visible as to outline; hymenial portion zonate, composed of 2 layers, each containing numerous curved, slender, flexuous, deeply staining organs  $30–75\times3–4$   $\mu$ , which may be elongated gloeocystidia or perhaps basidia of the transversely septate kind; a few scattered, brownish spherical organs resembling gloeocystidia of *Peniophora serialis*; spores hyaline, even,  $12–13\times4–5$   $\mu$ , few seen and may not belong.

On decaying logs. South Carolina.

In the original description it was stated that there is a thin, tomentose subiculum composed of interwoven threads. If so, it is not retained in my mounts of sections from the specimens in Kew and Farlow Herbaria made 26 and 24 years ago respectively. I did not decide from the type specimens whether this species is a Corticium or Stereum having elongated gloeocystidia or an Auricularia with transversely septate basidia. I noted the presence of the word "Auricularia" on the specimen in Kew Herbarium but the species was published as a Corticium. My thin Corticium argentatum is of too different structure to be a synonym of this. While writing this account it occurs to me that the specimens distributed in Ellis, N. Am. Fungi, 1109, under the name Phlebia spilomea, should have been compared with a type of C. venosum.

Specimens examined:

South Carolina: Black Oak, H. W. Ravenel, 1321, type (in Kew Herb. and in Farlow Herb.).

# 85. C. ochrofarctum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, hypochnoid, tilleul-buff in the herbarium, even, not shining, not cracked, the margin whitish, thinning out, with hyphae interwoven; in section  $100-150~\mu$  thick, not colored, composed of hyphae and numerous scattered, spherical, ochraceous gloeocystidia; hyphae rather

loosely arranged near the substratum, suberect, incrusted,  $4\frac{1}{2}\mu$  in diameter under the incrustation and up to  $6\mu$  over it, not incrusted and more densely arranged towards the hymenium; gloeocystidia in the form of brown or ochraceous, resinous, spherical or somewhat angular masses  $9-20\mu$  in diameter; spores white in a spore collection, even, cylindric, somewhat curved,  $8\times2\frac{1}{2}\mu$ .

Fructifications 2-6 cm. long, 5 mm.-3 cm. wide.

On decorticated, very rotten logs of *Populus trichocarpa*. Idaho. September.

The specific name ochrofarctum has reference to the colored, resinous gloeocystidia which are so large and so deep colored that they may be seen by inspection of the fructification with a lens and give, when so viewed, a minutely speckled appearance to the fructification. The large, coarsely incrusted hyphae are distinctive also. C. coroniferum is a related European species.

Specimens examined:

Idaho: Coolin, J. R. Weir, 11120, type, and 11122 (in Mo. Bot. Gard. Herb., 63695 and 63696 respectively).

# 86. C. Tsugae Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, thin, dry, hypochnoid, downy, avellaneous, with the surface white-pruinose, even, not cracked, the margin similar, indeterminate; in section 30  $\mu$  thick, not colored, composed of hyphae and numerous colored gloeocystidia; hyphae hyaline, thin-walled, 3  $\mu$  in diameter, not incrusted, not nodose-septate, ascending from the substratum; gloeocystidia in the form of brown or ochraceous, resinous-appearing, subspherical masses up to 18  $\mu$  in diameter; not more than 4 sterigmata to a basidium demonstrated; spores hyaline, even,  $6-7\frac{1}{2} \times 3-3\frac{1}{2} \mu$ , copious.

Fructifications in fragments up to  $2\frac{1}{2}$  cm. long,  $1\frac{1}{2}$  cm. wide. On very rotten wood of *Tsuga canadensis*. New Hampshire. September.

The color of the fructification of this species is so nearly that of the rotten substratum that close inspection is necessary to detect the presence of the fungus, whose color is probably due to the gloeocystidia. This aspect, together with uncommon gloeocystidia and non-incrusted hyphae, are good distinctive characters.

Specimens examined:

New Hampshire: Chocorua, W. G. Farlow, 148, type (in Mo. Bot. Gard. Herb., 55248).

### 87. C. subcinereum Burt, n. sp.

Type: in Burt Herb.

Fructifications long-effused, closely adnate, thin, not at all separable, pale gull-gray to pale drab-gray, slightly granular, somewhat pruinose, becoming cracked in drying, the margin similar or paler, thinning out; in section 60–100  $\mu$  thick, slightly colored, with the hyphae densely interwoven, 1–2  $\mu$  in diameter, so grown together as to show structure indistinctly, but probably not nodose-septate nor incrusted; no cystidia nor gloeocystidia; branched paraphyses about 1  $\mu$  in diameter are present in the hymenium; spores hyaline, even, 5–8  $\times$  3–3½  $\mu$ .

Fructifications 2-10 cm. long, 1-2 cm. wide.

On bark of fallen, decaying limbs of *Betula*, *Cornus*, and *Syringa*. Canada, Massachusetts, and Kansas. February to October. Local.

C. subcinereum closely resembles Peniophora cinerea, P. caesia, and C. argentea in aspect but is distinct from each by its lack of cystidia and gloeocystidia.

Specimens examined:

Canada: Ottawa, J. Macoun, 37, type.

Massachusetts: Sharon, A. P. D. Piguet, comm. by W. G. Farlow, 8 (in Mo. Bot. Gard. Herb., 55289).

Kansas: Rockport, E. Bartholomew; Rooks County, comm. by Lloyd Herb., 2301; Stockton, E. Bartholomew, 8620, 8702 (in Mo. Bot. Gard. Herb., 62491, 63749, and Burt Herb.).

88. C. albido-carneum (Schw.) Massee, Linn. Soc. Bot. Jour. 27: 142. 1890.

Thelephora albido-carnea Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 169. 1832.—Corticium albido-carneum (Schw.) Ravenel, Fungi Car. 4: 14, was a misdetermination by Ravenel.

Type: in Schweinitz Herb., Farlow Herb., and Kew Herb.

Fructifications effused, small, becoming confluent longitudinally but very narrow, closely adnate, thin, becoming pale drabgray to pinkish buff in the herbarium, pruinose, cracking transversely in drying, the margin paler; in section 60–120  $\mu$  thick, composed of 3 equal layers, of which that next to substratum consists of densely, longitudinally interwoven, slightly colored hyphae  $1\frac{1}{2}-2$   $\mu$  in diameter, not incrusted nor nodose-septate; the middle layer contains numerous pyriform bodies  $12 \times 6$   $\mu$  which are presumably basidia; the outer layer is composed of bushy-branched paraphyses 3  $\mu$  in diameter with final branchlets and lateral prongs about  $\frac{1}{2}$   $\mu$  in diameter; detached spores 5–8  $\times$  3–4  $\mu$ , few present and may not belong.

Fructifications 6-10 mm. long, 1-2 mm. wide, becoming more or less confluent over areas up to 5 cm. long and 3 cm. wide.

In crevices of the bark of dead wood of wild species of *Vitis*. Pennsylvania, Virginia, and Michigan. February and May. Rare.

C. albido-carneum is a very rare species which has been collected but few times and in small quantity for critical study. The specimens seem immature and the tissues of the fructifications are so minute and the covering of paraphyses so trouble-some that I have been unable to make out the detailed structure of the basidia. The plan of structure is suggestive of a Sebacina but I have been unable to demonstrate longitudinal septa in any of the pyriform organs. The somewhat smoky color of the sections, their 3-layered structure, and occurrence on bark of dead wild grape trunks are a combination of characters which should afford ready recognition of this species. The dates of collection of the specimens seem to indicate that the species may fruit in winter. If some of the pyriform organs are gloeocystidia, C. pilosum may prove not specifically distinct.

Specimens examined:

Pennsylvania: Bethlehem, Schweinitz, type (in Schweinitz Herb., Kew Herb., and Farlow Herb.).

Virginia: Arlington Farm, C. L. Shear, 2810 (in Mo. Bot. Gard. Herb., 15310).

Michigan: Paw Paw, L. A. Hawkins, comm. by C. L. Shear.

#### 89. C. adhaesum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, rather thick, not separable, between drab and deep olive-buff, somewhat granular, pulverulent, cracked at intervals of 1–2 mm., the margin abrupt; in section 250–350  $\mu$  thick, chamois-colored within, composed of densely arranged, thick-walled, erect and interwoven hyphae 3–3½  $\mu$  in diameter, not incrusted, not nodose-septate, conglutinate and not showing structure well; no gloeocystidia; spores hyaline, even, flattened on one side,  $3\frac{1}{2}-6 \times 2\frac{1}{2}-3 \mu$ , copious.

Fructifications 6 cm. long, 2 cm. wide.

On rough surface of badly decayed wood of a frondose species. Mexico and West Indies. Probably rare.

C. adhaesum is separated from the most of our species by having its fructifications colored within to such a degree that the thin sections are somewhat chamois-colored. The drab color of the hymenium and gluing together of the hyphae in sections are other distinctive characters.

Specimens examined:

-Mexico: Jalapa, W. A. & E. L. Murrill, 64, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 16479).

Porto Rico: Rio Piedras, J. A. Stevenson, 5577 (in Mo. Bot. Gard. Herb., 11059).

Jamaica: A. E. Wight, comm. by W. G. Farlow, C 1, type (in Mo. Bot. Gard. Herb., 44005).

Trinidad: Port of Spain, R. Thaxter, comm. by W. G. Farlow, 22. Grenada: Grand Etang, R. Thaxter, comm. by W. G. Farlow, 121.

C. leptaleum Ell. & Ev. in Millsp. & Nutt. Field Mus.
 Publ. Bot. 1: 170. 1896; Sacc. Syll. Fung. 14: 220. 1899.

Type: in N. Y. Bot. Gard. Herb.

Fructifications effused, adnate, membranaceous-soft, contracting in drying so that only one-half the original area is covered, cracking into masses 2–3 mm. in diameter and curling up from substratum so as to resemble cups of a *Peziza*, grayish white, becoming pinkish buff in the herbarium, pulverulent; in section  $300~\mu$  thick, composed of densely interwoven hyphae  $3-3\frac{1}{2}~\mu$  in diameter, incrusted in the subhymenium, only rarely nodose-septate; no gloeocystidia; spores hyaline, even,  $8-10~\times~3-4~\mu$ .

On under side of dead Magnolia Fraseri. West Virginia. April.

In the original description it is stated, "The membrane on which the hymenium stands where exposed on the incurved margin of the pezizoid areas is pale brown." Some twenty years ago at the time my sections of the type were made, I did not record whether the sections were colored within or not. They are now colorless but may have faded. The large spores preclude reference to C. hydnans.

Specimens examined:

West Virginia: L. W. Nuttall, 690, type (in N. Y. Bot. Gard. Herb.).

91. C. laeve Persoon, Roemer Neues Mag. Bot. 1:110. 1794; Sacc. Syll. Fung. 6: 611. 1888; Bresadola, Ann. Myc. 1: 94. 1903; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 232. 1911; Rea, Brit. Basid. 673. 1922.

Thelephora laevis Persoon, Syn. Fung. 575. 1801 (under Corticium); Myc. Eur. 1: 130. 1822.—T. evolvens Fries, Obs. Myc. 1: 254. pl. 4, f. 5. 1815; Syst. Myc. 1: 441. 1821; Elench. Fung. 1: 181. 1828.—Corticium evolvens Fries, Epicr. 557. 1838; Hym. Eur. 646. 1874; Sacc. Syll. Fung. 6: 604. 1888; Massee, Linn. Soc. Bot. Jour. 27: 118. pl. 6, f. 4. 1890.—Not Corticium laeve Fries, which is a Peniophora.

Type: in Herb. Mougeot, according to Bresadola in letter. Fragment of type from Quelet to Bresadola in Burt Herb.

Fructifications usually widely effused, rarely small and disk-shaped, very rarely slightly reflexed, thick, membranaceous, tender, small pieces separable when moist, becoming cream color and light pinkish cinnamon to wood-brown and drab in the herbarium, waxy, even, more or less undulate, sometimes coarsely tuberculate, cracking in drying and showing on the sides of the fissures a thick, crust-like hymenial layer of about the same color as the surface of the hymenium and connected with the substratum by a thicker layer of whitish floccose or loose tissue, the margin white, silky, radiating, but sometimes free when the fructifications are pezizaeform and 1–3 mm. in diameter; in section 200–500 µ thick, 2-layered, with the hymenial layer usually

somewhat colored but concolorous with the surface of the hymenium, very compact, supported by the broad layer of loosely arranged, obliquely ascending, thin-walled hyphae 3–4  $\mu$  in diameter, sometimes conspicuously guttulate, nodose-septate, not incrusted; no gloeocystidia; spores hyaline, even, 7–10  $\times$  4–6  $\mu$ , flattened on one side, tapering towards the pointed base, usually glued together on the flattened side at ends of the protruding basidia.

Fructifications 1-10 cm. long, 1-5 cm. wide, rarely only 1-3 mm. in diameter.

On bark of fallen decaying limbs of many frondose species. Europe and northern United States and Canada. Throughout the year. Very common.

C. laeve is a very common species on fallen limbs of poplar, maple, beech, etc., whose usually drab fructifications crack when dried and show the dark hymenial crust supported on a whitish subiculum. The absence of paraphyses and presence of spores  $7-10 \times 4-6 \,\mu$ , shaped like apple seeds and glued together in groups of 2-4, are important additional characters. In the large number of gatherings cited below there are only 2 American specimens which have a slightly reflexed margin and would be referred to Stereum, where the species really belongs.

Specimens examined:

Exsiccati: Brinkmann, Westfälische Pilze, 9; Cooke, Fungi Brit., 10; Ell. & Ev., Fungi Col., 221, under the name Corticium glabrum; Libert, Pl. Crypt. Ard., 20; Romell, Fungi Scand., 124; Sydow, Myc. Germ., 355, under the name Peniophora laevis; de Thümen, Myc. Univ., 1109.

Sweden: Svex. Söderm., Lindblad, authentic specimen of C. evolvens from Fries (in Kew Herb.); Stockholm, L. Romell, 89, 90, 91, 92, 93, 94, 95, and in Romell, Fungi Scand., 124.

Finland: Mustiala, P. A. Karsten, in de Thümen, Myc. Univ., 1109.

Germany: Brandenburg, H. Sydow, in Sydow, Myc. Germ., 355; Westphalia, W. Brinkmann, in Brinkmann, Westfälische Pilze, 9.

Austria: Innsbruck, Tirol, V. Litschauer, 3 specimens.

Italy: Trient, G. Bresadola, 3 specimens; Vallambrosa, Cavara, comm. by Bresadola.

- France: A. Libert, in Libert, Pl. Crypt. Ard., 20; Paris, Persoon, original specimen of C. laeve, comm. by Bresadola; Strassburg, L. Maire.
- England: E. M. Wakefield (in Mo. Bot. Gard. Herb., 58691); Hampstead, in Cooke, Fungi Brit., 10.
- Canada: Lower St. Lawrence Valley, J. Macoun, 17, 50.
- Ontario: Granton, J. Dearness, 1040 E (in Mo. Bot. Gard. Herb., 23107); London, J. Dearness, 945 h (in Mo. Bot. Gard. Herb., 14252).
- Newfoundland: Bay of Islands, A. C. Waghorne, 517, 1027.
- New Hampshire: Chocorua, W. G. Farlow, 2 (in Mo. Bot. Gard. Herb., 44594).
- Vermont: Middlebury, E. A. Burt, 5 gatherings, Ripton, E. A. Burt, 4 gatherings.
- New York: Adirondack Mts., G. F. Atkinson, C; Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 14829, 19456); Alcove, C. L. Shear, 1210, 1214; Altamont, E. A. Burt; Bronx Park, Class in Mycology (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61389); Hague, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 56110), and 13; Ithaca, C. O. Smith, comm. by G. F. Atkinson, 8046, and G. F. Atkinson, d, 2813, 4899; Lyndonville, C. E. Fairman, 138 (in Mo. Bot. Gard. Herb., 61438); New York, F. S. Earle (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61677); Newcomb, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 59666); Oneida, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 59679, 59699); Sylvan Beach, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 7461); Syracuse, L. M. Underwood, 51, 126 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61571, 61394), and in Ell. & Ev., Fungi Col., 221.
- District of Columbia: Takoma Park, C. L. Shear, 1038.
- Michigan: Michigan Agricultural College, B. O. Longyear, 9 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55787).
- Missouri: St. Louis, E. A. Burt (in Mo. Bot. Gard. Herb., 58334).
- British Columbia: Sidney, J. Macoun, 65, 77, 78 (in Mo. Bot. Gard. Herb., 5743, 5753, 9778), and 35, 288, 319, 350, 424 (in Macoun Herb.): Squamish, J. Macoun, 318, 536 (in Mo. Bot.

Gard. Herb.); Victoria, J. Macoun, 577 (in Macoun Herb.); Vancouver Island, J. Macoun, 419 (in Mo. Bot. Gard. Herb., 55315), and comm. by J. Dearness, V 35 (in Mo. Bot. Gard. Herb., 19573).

Washington: Bingen, W. N. Suksdorf, 714, 755, 872, 886, 898, 899, 901, 955, 961; Olympia, C. J. Humphrey, 6293, 6330;
Seattle, A. M. Parker, 177 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61475).

Oregon: Seattle, W. A. Murrill, 988, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 55703).

92. C. investiens (Schw.) Bresadola, I. R. Accad. Agiati Atti III. 3: 110. 1897; Ann. Myc. 1: 95. 1903.

Radulum? investiens Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 165. 1832; Sacc. Syll. Fung. 11: 112. 1895.—Vararia investiens (Schw.) Karsten, Krit. Öfvers. Finl. Basidsv. Tilläg 3: 32. 1898.—Asterostromella investiens (Schw.) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 117: 1083. 1908.—Corticium alutarium Berk. & Curtis, Grevillea 2: 4. 1873; Sacc. Syll. Fung. 6: 634. 1888; Massee, Linn. Soc. Bot. Jour. 27: 137. 1890.—Thelephora subochracea Peck, N. Y. State Mus. Rept. 46: 109. 1893; Sacc. Syll. Fung. 11: 116. 1895.—Xerocarpus alutarius (Berk. & Curtis) Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 48: 418. 1889.

Type: in Schweinitz Herb., Farlow Herb., Fries Herb., and probably in Kew Herb.

Fructifications broadly effused, usually thin, tough, dry, adnate, small pieces separable when moist, warm buff to light orange-yellow, conforming to inequalities of the substratum, somewhat tomentose, not cracked, the margin thinning out; in section 150–600  $\mu$  thick, concolorous with the hymenium, composed of a few even-walled, hyaline hyphae  $21/2~\mu$  in diameter, and of a great number of yellowish, stiff hyphae with dichotomous and antler-shaped branching and short, acicular, prong-like terminal branchlets, which extend beyond the basidia in the hymenial surface; no gloeocystidia; basidia 4-spored; spores hyaline under the microscope but slightly straw-colored in the mass, even,  $12\times 4~\mu$ , tapering downward to the slender, apiculate base.

Fructifications 2-20 cm. long, 1-5 cm. wide.

On rotten logs and fallen branches of both frondose and coniferous species and sometimes running over fallen leaves and the ground. In Europe, throughout North America, West Indies, Venezuela, and in Japan. July to December. Very common.

C. investiens is readily recognized by chamois color and surface texture like that of chamois leather. Under the microscope the antler-shaped branching of its principal hyphal component is well shown. This mode of hyphal branching seems to me a useful specific character for the various other species which have it, e. g., Lachnocladium brasiliense, Grandinia granulosa, Stereum induratum, S. duriusculum, Hypochnus peniophoroides, H. pallescens, Peniophora phyllophila, P. piliseta, P. mexicana, and Corticium jamaicense but not of greater importance than other hyphal modifications which are useful specific characters, hence I can not accept as helpful Karsten's genus Vararia, of which the type species is Corticium investiens, nor its synonym Asterostromella of v. Höhnel & Litschauer.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 517.

Sweden: Femsjö, L. Romell, 157, and C. G. Lloyd, 09149 (in Mo. Bot. Gard. Herb., 55619).

Hungary: Kmet, comm. by Bresadola.

Canada: J. Macoun, 91.

Ontario: Niagara, J. Dearness, D586 (in Mo. Bot. Gard. Herb., 3727); Temagami, C. G. Lloyd, 07633 (in Mo. Bot. Gard. Herb., 55618).

Maine: Kittery Point, R. Thaxter & E. A. Burt.

New Hampshire: Chocorua, W. G. Farlow; Shelburne, W. G. Farlow.

Vermont: Grand View Mt., E. A. Burt, 2 gatherings; Lake Dunmore, E. A. Burt; Little Notch, E. A. Burt; Middlebury, E. A. Burt.

Massachusetts: Lincoln, A. B. Seymour, T40 (in Mo. Bot. Gard. Herb., 12955); Magnolia, W. G. Farlow (in Farlow Herb.).

New York: Albany, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 6324, 54358, 54359); Alcove, C. L. Shear, 1121, 1123, 1203, 1322; Arkville, W. A. Murrill (in N.

Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61361); East Galway, E. A. Burt; Floodwood, E. A. Burt, C. H. Peck, 4a; Fort Ann, S. H. Burnham, 25 (in Mo. Bot. Gard. Herb., 54495); Freeville, G. F. Atkinson, 2812; Gansevoort, C. H. Peck (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 55974); Ithaca, G. F. Atkinson, 8200, 22758, 22763, 23278, and C. J. Humphrey, 548, 22563; Karner, H. D. House, 14,154, comm. by N. Y. State Mus. Herb. (in Mo. Bot. Gard. Herb., 44711); Lake Placid, W. A. & E. L. Murrill, 282 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 61673); North Elba, C. H. Kauffman, 6 (in Mo. Bot. Gard. Herb., 21464); North Greenbush, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55783, 56109); Oneida, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57474, 59682); Onondaga Valley, L. M. Underwood, 11 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61580); Sandlake, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55779); Shokan, C. H. Peck, type of Thelephora subochracea (in N. Y. State Mus. Herb.); Snyders, C. H. Peck (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55780); Westport, C. H. Peck, 4.

Pennsylvania: Michener, type of Corticium alutarium (in Curtis Herb., 6349); Bethlehem, Schweinitz, type of Radulum? investiens (in Schweinitz Herb. and Farlow Herb.) and under the name Thelephora ochracea of Schweinitz (in Curtis Herb. from Schweinitz Herb.); Ohio Pyle, W. A. Murrill, 1047 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61553); State College, L. O. Overholts (in Mo. Bot. Gard. Herb., 54701); Trexlertown, W. Herbst, 33, 42; West Chester, Everhart & Haines, in Ell. N. Am. Fungi, 517; no locality given, H. Jackson, Gentry (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55809, 55905, respectively).

Delaware: Newark, H. S. Jackson, B7.

District of Columbia: Takoma Park, C. L. Shear, 960.

Louisiana: St. Martinville, C. J. Humphrey, 2519 (in Mo. Bot. Gard. Herb., 42937).

West Virginia: Eglon, C. G. Lloyd, 1408 (in Mo. Bot. Gard. Herb., 55610); Nuttallburg, L. W. Nuttall, 189, comm. by U. S. Dept. Agr. Herb.; Paw Paw, C. L. Shear, 1172.

- Ohio: C. G. Lloyd, 4197 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61593); Cincinnati, A. P. Morgan, the Corticium ochraceum of Morgan Herb., comm. by Lloyd Herb., 2639.
- Indiana: Millers, E. T. & S. A. Harper, 830.
- Michigan: Ann Arbor, C. H. Kauffman, 41 (in Mo. Bot. Gard. Herb., 22930); Whitmore Lake, A. H. W. Povah, 10 (in Mo. Bot. Gard. Herb., 9228).
- Montana: Trego, E. E. Hubert, comm. by J. R. Weir, 12039 (in Mo. Bot. Gard. Herb., 63389).
- Idaho: Priest River, J. R. Weir, 38; E. E. Hubert, comm. by J. R. Weir, 11998 (in Mo. Bot. Gard. Herb., 63361).
- British Columbia: Sidney, J. Macoun, 14 (in Mo. Bot. Gard. Herb., 5732); Vancouver Island, J. Macoun, comm. by J. Dearness, V148 (in Mo. Bot. Gard. Herb., 21138).
- Mexico: Orizaba, Barrio Nuevo, W. A. & E. L. Murrill, 762, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54646).
- Jamaica: Castleton Gardens, W. A. & E. L. Murrill, 123 (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., 61365, and Burt Herb.); Cinchona, W. A. & E. L. Murrill, 648 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61458); Morces Gap, W. A. & E. L. Murrill, 734 (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61466).
- Porto Rico: Rio Piedras, J. A. Stevenson, 3474 (in Mo. Bot. Gard. Herb., 6732).
- Venezuela: Fendler (in Curtis Herb., 190, under the herbarium name Corticium xanthellum).
- Japan: Nakada-mura, Prov. Awaji, A. Yasuda, 44 (in Mo. Bot. Gard. Herb., 56169).

# 93. C. pectinatum Burt, n. sp.

Type: in Mo. Bot. Gard. Herb. and Farlow Herb.

Fructifications broadly effused, thin, closely adnate, not separable, warm buff to wood-brown in the herbarium, cracking into polygonal masses about 2 to the mm., not shining, the margin unknown; in section 60–90  $\mu$  thick, concolorous with the hymenium, composed of densely interwoven, colorless or slightly colored hyphae about 1  $\mu$  in diameter, not incrusted, not nodose-

septate, and of comb-shaped or antler-shaped branching, slightly colored masses of about 5–10  $\mu$  in diameter each and having many prongs; no gloeocystidia; basidia 6–12  $\times$  4–5  $\mu$ , immature, immersed in the antler-shaped paraphyses which form the surface of the hymenium; no spores found.

Fructification 1-6 cm. long, \frac{1}{2}-1\frac{1}{2} cm. wide.

On bark of dead frondose limbs. Florida and West Indies. October to March.

C. pectinatum has the general aspect and color of C. scutellare and structure of C. investiens but with much smaller and more delicate hyphae and antier-shaped organs than the latter.

Specimens examined:

Florida: Cocoanut Grove, R. Thaxter, 76, type (in Mo. Bot. Gard. Herb., 43898); Royal Palm Hammock, W. A. Murrill, 131, comm. by N. Y. Bot. Gard. Herb., 63762).

Cuba: Omaja, C. J. Humphrey, 2596 (in Mo. Bot. Gard. Herb., 8730).

#### 94. C. racemosum Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, closely adnate, thin, dry, not separable, drying cream-buff, even, not shining, becoming transversely cracked in the central portions, the margin thinning out, indeterminate, concolorous; in section 70–140  $\mu$  thick, colored cream-buff, composed of very densely arranged, erect, branching and interwoven hyphae 2–2½  $\mu$  in diameter; no gloeocystidia; paraphyses in hymenial surface with tips branched sometimes racemosely, sometimes in antler-shaped manner, often irregularly, these branches about ½  $\mu$  in diameter; spores hyaline, even, flattened on one side, 4–6  $\times$  2–3  $\mu$ .

Fructifications 2-12 cm. long, 1-4 cm. wide.

On bark and wood of decaying logs of Thuja plicata, Larix occidentalis, Abies grandis, and Pseudotsuga taxifolia. Idaho, British Columbia, and Washington. July to September.

The slender branched paraphyses of *C. racemosum* and lack of gloeocystidia locate this species in the group with *C. Atkinsonii*, *C. albidocarneum*, *C. rubropallens*, and *C. rubrocanum*. The antlershaped branching of occasional paraphyses connects this species

with the C. investiens group also. Radulum Pini-canadense Schw. should also be considered here.

Specimens examined:

Idaho: Priest River, J. R. Weir, 39, type, and 137 (in Mo. Bot. Gard. Herb., 9852).

British Columbia: Salmo, J. R. Weir, 465 (in Mo. Bot. Gard. Herb., 11777).

Washington: Stanwood, C. J. Humphrey, 7360 (in Mo. Bot. Gard. Herb., 7825).

95. C. subcontinuum Berk. & Curtis, Linn. Soc. Bot. Jour. 10: 337. 1868; Sacc. Syll. Fung. 6: 635. 1888; Massee, Linn. Soc. Bot. Jour. 27: 128. 1890.

Type: in Kew Herb. and Farlow Herb.

Fructifications effused, adnate, rather thick, small pieces separable, becoming chamois-colored in the herbarium, ceraceous, even, sometimes cracking in drying but the cracks not running together, showing the Isabella-colored tissue on the sides of the cracks, the margin thinning out; in section 200–400  $\mu$  thick, Isabella-colored, 2-layered, with a broad layer next to the substratum of brown hyphae 2-3  $\mu$  in diameter, not incrusted, not nodose-septate; spores hyaline, even, subglobose, 3–4  $\mu$  in diameter or 4  $\times$  3  $\mu$ .

Fructifications recorded as "spreading for several inches." The fragmentary pieces in herbaria are 2-3 cm. long, 1 cm. wide.

On bark and decaying wood. Louisiana, Texas, and West Indies. February to June. Rare.

The fructifications of C. subcontinuum resemble in general aspect, thickness, and consistency those of C. confluens, but are of different structure from those of the latter and are sharply distinct by the colored substance of the interior. The Louisiana specimens are doubtfully referred to this species.

Specimens examined:

Louisiana: Ruston, C. J. Humphrey, 2532 (in Mo. Bot. Gard. Herb., 12495); St. Martinville, A. B. Langlois, 1761 b, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 42598, and Burt Herb.) and 1761 a, in part.

Texas: locality not given, C. Wright, comm. by U. S. Dept. Agr. Herb., under the name C. calceum.

Cuba: C. Wright, 537, type (in Kew Herb. and Curtis Herb.); Omaja, C. J. Humphrey, 2575.

Porto Rico: Rio Piedras, J. A. Stevenson & R. C. Rose, 6528 (in Mo. Bot. Gard. Herb., 55083).

#### 96. C. Murrilli Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications broadly effused, thick, soft, spongy, dry, flexible, separable in sheets which have the hymenium between light buff and cream-buff and the under side Van Dyke brown, hymenium velutinous, not cracked, the margin concolorous with the hymenium, tomentose; in section 600–900  $\mu$  thick, colored, with the hyphae of the under layer brown, loosely, longitudinally interwoven, rough, nodose-septate,  $4-4\frac{1}{2}\mu$  in diameter, and with hymenial layer 75–450  $\mu$  thick with hyaline, interwoven hyphae; no gloeocystidia; basidia simple, with 4 sterigmata; spores hyaline, even, cylindric, 25–35  $\times$  6–9  $\mu$ .

Fructification 7 cm. long,  $3\frac{1}{2}$  cm. wide in the piece seen which is broken off at one end and on one side.

On bark of decaying log of an apparently frondose species in a moist virgin forest. Mexico. December.

C. Murrilli is probably a species with large, soft, dry fructification separable from the bark in a pliant, sheet-like mass and having the hymenium buff color and the under side a rich Van Dyke brown. The very large spores are another distinguishing character. C. Langloisii is thinner and has smaller spores.

Specimens examined:

Mexico: Jalapa, W. A. & E. L. Murrill, 182, type, comm. by N. Y. Bot. Gard. (in Mo. Bot. Gard. Herb., 44967).

97. C. subochraceum Bresadola, Hedwigia 35: 290. 1896; Sacc. Syll. Fung. 14: 221. 1899.

Type: part of type in Burt Herb.

Fructifications broadly effused, closely adnate, very thin, not separable, becoming light pinkish cinnamon to wood-brown in the herbarium, glabrous, even, not shining, not cracking, the margin thinning out, whitish at first, becoming colored like the hymenium; in section 45–100  $\mu$  thick, only slightly colored in the hy-

menium and subhymenium but giving the color to the fructification, composed of densely interwoven, distinct hyphae  $3-3\frac{1}{2}\mu$  in diameter, not incrusted, not nodose-septate; no gloeocystidia; spores hyaline, even,  $3-4\frac{1}{2}\times 2-2\frac{1}{2}\mu$ , copious.

Fructifications 1-8 cm. long, 1-2 cm. wide.

On bark and decaying wood of frondose species. Alabama, Louisiana, Nebraska, and Brazil. May and June.

C. subochraceum occurs on decaying frondose wood and bark in closely adnate, thin fructifications of wood-brown color due to the pale color of the superficial tissue. The spores were published by Bresadola as  $6-8 \times 4-4\frac{1}{2}\mu$  and the hyphae as conglutinate, but in the original specimen from Bresadola the spores are copious, flattened on one side, and not larger than  $4\frac{1}{2} \times 2\frac{1}{2}\mu$  and the hyphae not conglutinate.

Specimens examined:

Alabama: Auburn, Earle & Baker (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 58325); Montgomery Co., R. P. Burke, 338 (in Mo. Bot. Gard. Herb., 57212).

Louisiana: St. Martinville, A. B. Langlois, ab, w, and 1345, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 42603).

Nebraska: Lincoln, C. L. Shear, 1341.

Brazil: Blumenau, A. Möller, part of type from Bresadola.

## 98. C. canadense Burt, n. sp.

Type: in Burt Herb., Mo. Bot. Gard. Herb., and N. Y. State Mus. Herb.

Fructifications broadly effused, adnate, rather thick, membranaceous, small pieces separable when moistened, light buff, even, ceraceous, cracking but little in drying, the margin narrow, sulphur-yellow, with its hyphae interwoven; in section 600–800  $\mu$  thick, colored, stratose, the buried strata becoming fuscous; hyphae of each stratum 3  $\mu$  in diameter, not incrusted, occasionally nodose-septate, erect, loosely arranged below, forming a compact hymenium; no gloeocystidia; spores white in spore collection, cylindric, even,  $4\frac{1}{2}-6 \times 1\frac{1}{2}-2 \mu$ .

Fructifications 3-10 cm. long, 1-5 cm. wide.

On decaying wood of logs of *Pinus Strobus*. Canada and New Hampshire. July to September. Rare.

C. canadense has beautiful fructifications with buff hymenium and sulphur-colored margin. The occurrence on pine, stratose structure in section, and the buried strata fuscous in color afford more ample confirmatory distinctive characters than we usually find in resupinate species.

Specimens examined:

Canada: Ontario, Ottawa, J. Macoun, 26, type (in Burt Herb., N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55909).

New Hampshire: Chocorua, W. G. Farlow (in Mo. Bot. Gard. Herb., 6766), 8, and E. A. Burt.

99. C. bicolor Peck, Buffalo Soc. Nat. Hist. Bul. 1:62. 1873;
N. Y. State Mus. Rept. 26:72. 1874; Sacc. Syll. Fung. 6:630. 1888; Massee, Linn. Soc. Bot. Jour. 27:157. 1890.

Type: in N. Y. State Mus. Herb.

Fructifications widely effused, thin, membranaceous, tender, small pieces separable when moist, white, becoming pale pinkish buff to cream color in the herbarium, even, continuous, not cracked, the subiculum wax-yellow throughout, byssoid, the margin yellow to wax-yellow, often running out into wax-yellow rhizomorphic strands; in section 200–300  $\mu$  thick, yellow near the substratum and usually throughout, color not changed by lactic acid but bleached by potassium hydrate solution; the hyphae loosely interwoven, delicate,  $2\frac{1}{2}\mu$  in diameter, somewhat rough or incrusted with small crystals; no gloeocystidia; spores hyaline, even, subglobose,  $2\mu$  in diameter or  $3\times 2\mu$ , copious.

Fructifications 3-8 cm. long, 2-3 cm. wide.

On under side of fallen limbs and decaying wood on the ground, usually on pine and other conifers but also on *Populus*. New Hampshire to New Jersey and in Montana and Washington. August to November. Uncommon.

C. bicolor is a beautiful species related to C. sulphureum, from which it constantly differs in occurring nearly always in fertile condition with a compact whitish, even hymenium borne on the brilliant, wax-yellow subiculum. The hyphae and spores are similar to those of C. sulphureum.

Specimens examined:

New Hampshire: Chocorua, A. P. D. Piguet, comm. by W. G.

Farlow, 176, and W. G. Farlow (in Mo. Bot. Gard. Herb., 55249 and 13630, respectively).

New York: Karner, H. D. House, comm. by N. Y. State Mus. Herb., 14.152; Oneida, H. D. House (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 57452, 57476); Warrensburg, C. H. Peck, type (in N. Y. State Mus. Herb.) and (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 55771).

New Jersey: Newfield, J. B. Ellis, 88, comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 7944).

Montana: Evaro, J. R. Weir, 419, 435 (in Mo. Bot. Gard. Herb., 14768, 6707).

Washington: Hoquiam, C. J. Humphrey, 6400.

100. C. koleroga (Cooke) v. Höhnel, K. Akad. Wiss. Wien Sitzungsber. 119: 395. 1910; Burt, Mo. Bot. Gard. Ann. 5: 123. f. 1. 1918.

Pellicularia koleroga Cooke, Grevillea 4: 116, 134. 1876; Pop. Sci. Rev. 15: 164. pl. 135, f. a-c. 1876; Linn. Soc. Bot. Jour. 18: 461. 1881; Sacc. Syll. Fung. 4: 149. 1886; Fawcett, G. L., Porto Rico Agr. Exp. Sta. Ann. Rept. 1910: 35. 1911; Jour. Agr. Res. 2: 231. text f. 1-3. 1914; Porto Rico Agr. Exp. Sta. Bul. 17: 8. pl. 1. 1915.—Erysiphe scandens Ernst, A., Estudios sobre las Deformaciones, Enfermedades y Enemigos del Arbol de Cafe in Venezuela, 16. pl. f. 5. 1878.

Type: in Kew Herb.

The parasitic vegetative mycelium forms long, slender, mycelial strands of rather uniform diameter, whitish or pallid at first, finally fuscous, running along the branches and midrib and veins of the leaves, infecting the leaves and ramifying between the cells of the leaf parenchyma, finally emerging at many points on the under side of the leaf to form minute fructifications which give a mottled appearance to the leaf; fructifications soon laterally confluent into a thin, arachnoid, perforate membrane covering the under surface of the leaf between midrib and principal veins, drying pale smoke-gray, separable in small pieces, composed of loosely interwoven, hyaline or slightly colored, thin-walled, even, rigid hyphae  $41/2-6~\mu$  in diameter, not nodose-septate, running parallel with the substratum, and about 1–3 hyphae thick, branching at right angles; basidia scattered along the hyphae, simple, ovoid, 10–

 $12 \times 7-8 \,\mu$ , with short sterigmata; spores hyaline, even, flattened or slightly concave on one side,  $10-13 \times 3\frac{1}{2}-5 \,\mu$ .

Mycelial strands in the specimens received are 35 cm. long and broken with the branch at the lower end,  $\frac{1}{2}$ -1 mm. in diameter, not swollen into sclerotia; fructifications 9 cm. long, 4 cm. broad, 30–45  $\mu$  thick, more or less divided by the midrib and principal veins.

Parasitic on branches and leaves of the coffee plant. India, and the Antilles and neighboring regions of South America.

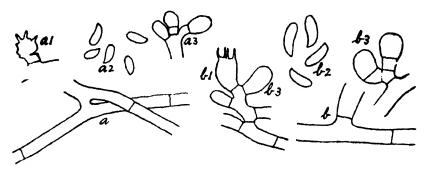


Fig. 1. C. koleroga. a-a3, from sketches by Miss Wakefield of structure of type in Kew Herbarium; magnification not stated but computed from spore dimensions at about 630. a, hypha; a1, collapsed basidium; a2, spores; a3, young basidia. b-b3, from Porto Rican specimen,  $\times$  870. b, hypha; b1, basidium; b2, spores; b3, young basidia.

Specimens examined:

India: Mysore, preparation from the type (in Kew Herb.).

Porto Rico: Mayaguez, F. L. Stevens, 9488 (in Stevens Herb., and in Mo. Bot. Gard. Herb., 54510); H. E. Thomas (in Mo. Bot. Gard. Herb., 55397).

Colombia: H. T. Dawe, fragment (in Mo. Bot. Gard. Herb. from specimen in Kew Herb.).

Venezuela: A. Ernst, fragments showing mottled stage and continuous fructification respectively (in Mo. Bot. Gard. Herb. from specimens in Kew Herb., determined by Ernst as Candelillo, Erysiphe scandens); H. Peltier, comm. by U. S. Dept. Agr., Path. & Myc. Coll., 1713 (in Mo. Bot. Gard. Herb., 62168).

**101.** C. Stevensii Burt, Mo. Bot. Gard. Ann. 5: 125. text f. 2. 1918.

Hypochnopsis ochroleuca Noack, Boletim do Instituto Agronomico Sao Paulo em Campinas 9: 80. 1898.—Hypochnus ochroleucus Noack in Sacc. Syll. Fung. 16: 197. 1902; Stevens, Science N. S. 26: 724. 1907; Stevens & Hall, Ann. Myc. 7: 49-59. text f. 1-8. 1909.—Not Corticium ochroleucum Bresadola, Fungi Trid. 2: 58. pl. 167, f. 2. 1892.

Vegetative mycelium forms on the twigs roundish or oblong, chestnut-brown sclerotia 3-4 mm. in diameter, and also slender mycelial strands white when young, becoming chestnut-brown, running along the twigs and petioles to the leaves and fructifying there; fructifications at first downy and barely visible, soon thickening into a dirty pinkish buff, felty membrane covering the whole under side of the leaf and frequently separable from it as a

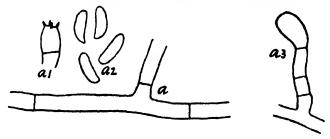


Fig. 2. C. Stevensii. From specimen from Trinidad, × 870. a, hypha; a1, basidium; a2, spores; a3, young basidium.

whole by mere handling; hyphae hyaline or slightly colored, giving their color to the fructifications, even, thin-walled, not incrusted, not nodose-septate,  $4\frac{1}{2}-7\frac{1}{2}\mu$  in diameter; basidia scattered along the hyphae on short lateral branches, simple,  $11 \times 7-8 \mu$ , with four short sterigmata; spores hyaline, flattened or slightly concave on one side,  $8-11 \times 3-4 \mu$ .

Fructification 11 cm. long, 3-4 cm. broad, 45-60  $\mu$  thick, unbroken over whole under surface of leaves; sclerotia 3-4 mm. in diameter; mycelial strands  $\frac{1}{2}$ -1 mm. in diameter, many cm. long.

On apple, pear, and quince, in Brazil and southern United States, causing the leaves to dry and fall, and on *Codiaeum* in Trinidad.

This species differs from *Corticium koleroga* by having sclerotia and thicker, darker-colored, and more felted fructifications which are but feebly attached to the leaf and form an unbroken covering over the whole under surface of the leaf from margin to margin. Fruiting specimens of this fungus have been available for study from only two localities, but these specimens agree in the characters stated above.

Specimens examined:

North Carolina: Horseshoe, J. G. Hall, comm. by F. L. Stevens, sclerotial stage on pear twigs; Mt. Airy, F. C. Reimer, comm. by F. L. Stevens, fertile stage on pear leaves.

Georgia: A. L. Quaintance, comm. by F. S. Earle, sclerotial stage on apple twigs.

Florida: C. G. Lloyd, sclerotial stage on pear twigs.

Texas: Dickson, F. W. Mally, comm. by U. S. Dept. Agr., sclerotial stage on pear twigs.

Trinidad: Diego Martei, J. B. Rorer, fertile stage on leaves of Codiaeum variegatum (in Mo. Bot. Gard. Herb., 44771); Petit Valley, J. B. Rorer, sclerotial and fruiting stages on leafy twigs of Codiaeum variegatum (in Mo. Bot. Gard. Herb., 11960, 19786, 19810, and 20062).

102. C. vagum Berk. & Curtis, Grevillea 1: 179. 1873; Sacc. Syll. Fung. 6: 616. 1888; Massee, Linn. Soc. Bot. Jour. 27: 148. 1890; Duggar, Mo. Bot. Gard. Ann. 2: 445. 1915; Peltier, Univ. Ill. Agr. Exp. Sta. Bul. 189: 285. 1915; Burt, Mo. Bot. Gard. Ann. 5: 128. text f. 3. 1918; Coker, Elisha Mitchell Scientif. Soc. Jour. 36: 173. pl. 33, f. 9, 10. 1921.

Corticium vagum Berk. & Curtis var. Solani Burt in Rolfs, Science N. S. 18: 729. 1903; Colo. Agr. Exp. Sta. Bul. 91: 1-20. pl. 1-5. 1904.—Hypochnus Solani Prill. & Del. Soc. Myc. Fr. Bul. 7: 220. text f. 1891; Sacc. Syll. Fung. 11: 130. 1895.—Corticium Solani Prill. & Del. in Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 248. 1911.—Corticium botryosum Bresadola, Ann. Myc. 1: 99. 1903; Sacc. Syll. Fung. 17: 173. 1905; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 248. 1911.—Rhizoctonia Solani Kühn, Krankheiten d. Kulturgewächse, 224. 1858; Duggar, Mo. Bot. Gard. Ann. 2: 424. 1915.

Type: in Kew Herb. and in Curtis Herb.

Vegetative mycelium saprophytic in the soil and in wood in contact with the ground, and parasitic as the Rhizoctonia Solani stage

in underground portions of various plants and forming at their surface underground minute sclerotia; fructification a thin, arachnoid, perforate membrane more or less separable, pale olive-buff to cream color; in structure 60–100  $\mu$  thick, composed of a few loosely interwoven hyphae running along the substratum and sending out short branches which bear the basidia; hyphae in contact with substratum may be slightly brownish, hyaline elsewhere, not incrusted, not nodose-septate, up to 6–10  $\mu$  in diameter, with branches smaller; basidia not forming a compact hymenium,  $10-20 \times 7\frac{1}{2}-11$   $\mu$ , with 4–6 sterigmata 6–10  $\mu$  long

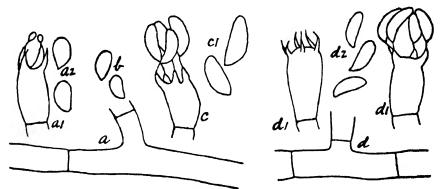


Fig. 3. C. vagum,  $\times$  870. a-al, from specimen on potato in Colorado. a, hypha; a1, basidium; a2, spores. b, spores of specimen on Plantago in Illinois. c-c1, from specimen on earth in Massachusetts. c, basidium; c1, spores. d-d2, from specimen on wood in British Columbia. d, hypha; d1, basidia; d2, spores.

and more or less swollen towards the basidium; spores hyaline, even, flattened on one side,  $8-14 \times 4-6 \mu$ .

Fructifications 5-15 cm. long on logs, 5-10 cm. broad; in a collar 1-10 cm. long, sheathing the base of living stems.

On bare earth, wood and bark lying on the ground, and on living stems of potatoes, beans, rhubarb, horseradish, tomatoes, Amaranthus, etc., at or near the ground. New Brunswick to Florida and westward to Vancouver and Washington, in West Indies, Europe, India, and Australia. Common.

Corticium vagum differs from C. koleroga and C. Stevensii in having its mycelium and sclerotia subterranean when parasitic, in having its fructifications at the surface of the ground or merely sheathing small herbaceous stems for only a few centimeters up from the ground and never spreading out on the under side of

broad leaves at a considerable distance above ground, by having larger hyphae, larger basidia, and the basidia with larger sterigmata which are more thickened in the lower portion and sometimes six to a basidium; the spores are somewhat larger in *C. vagum* also. The examination of the large amount of *C. vagum* which has come to hand does not afford ground for regarding the collar-like fructifications on small living herbaceous stems as worthy of varietal separation. As common as this species now is in the United States, it is rather surprising that a collection of it under some name has not been found in Herb. Schweinitz.

Specimens examined:

Exsiccati: Ellis, N. Am. Fungi, 330; Ravenel, Fungi Am., 132, 577—the latter under the name Zygodesmus pannosus.

Sweden: Stockholm, L. Romell, 204.

Russian Poland: Eichler, comm. by Bresadola, portion of type of Corticium botryosum Bres.

New Brunswick: Campobello, W. G. Farlow, 3.

Canada: J. Macoun, 2, 84, 340.

Ontario: Ottawa, J. Macoun, 327.

Massachusetts: Brookline, G. R. Lyman, 180; Magnolia, W. G. Farlow.

New York: Albany, H. D. House & J. Rubinger (in Mo. Bot. Gard. Herb., 8734); East Galway, E. A. Burt, 2 collections; Ithaca, Van Hook, comm. by G. F. Atkinson, 8092; Karner, H. D. House, 14.162, and 3 other collections (in N. Y. State Herb. and Mo. Bot. Gard. Herb., 44709, 54349, 55199, 55203); Tripoli, S. H. Burnham, 13, in part (in Mo. Bot. Gard. Herb., 54506).

New Jersey: Belleplain, C. L. Shear, 1244; Newfield, J. B. Ellis, in Ellis, N. Am. Fungi, 330.

Pennsylvania: Carbondale, E. A. Burt; Trexlertown, W. Herbst, 95.

Maryland: Takoma Park, C. L. Shear, 1164, 1334.

District of Columbia: Takoma Park, C. L. Shear, 965, 1041 (the former in Mo. Bot. Gard. Herb. also).

South Carolina: Curtis Herb., 3240, type (in Kew Herb. and in Curtis Herb.); Aiken, H. W. Ravenel, in Ravenel, Fungi Am., 132, 577.

Alabama: Montgomery, R. P. Burke, 170 (in Mo. Bot. Gard. Herb., 43162).

West Virginia: Paw Paw, C. L. Shear, 1171.

Ohio: Cincinnati, C. G. Lloyd, 4508.

Illinois: Urbana, G. L. Peltier, 14 collections, on living stems of beans, carrot, tomato, radish, rhubarb, horseradish, potato, winter vetch, spinach, Amaranthus, Campanula, and Plantago major (in Mo. Bot. Gard. Herb., 6264, 8761-8765, 8816, 43836, 44677-44682).

Montana: Evaro, J. R. Weir, 434 (in Mo. Bot. Gard. Herb., 17725).

Idaho: Coolin, J. R. Weir, 11545 (in Mo. Bot. Gard. Herb., 63298). Priest River, J. R. Weir, 140, 89 in part (Mo. Bot. Gard. Herb., 8197, 11349).

Colorado: Fort Worth, F. M. Rolfs, 2 collections, on living stems of potatoes.

Manitoba: Norway House, G. R. Bisby, 1475, 1477 (in Mo. Bot. Gard. Herb., 61657, 61659).

British Columbia: Sidney, J. Macoun, 4, 20, 83, 85, 87, 26, 154 (in Mo. Bot. Gard. Herb., 5764, 5735, 7068, 7024, 7833, 55347, 55350, respectively) and 39a, 151, 172 (in Macoun Herb.); Vancouver Island, J. Macoun, V89, V90, V151, V154, V172 (in Mo. Bot. Gard. Herb., 22815, 22927, 20357, 20507, 20728, respectively).

Washington: Bingen, W. N. Suksdorf, 846, 852, 863.

India: Ceylon, T. Petch, 5675 (in Mo. Bot. Gard. Herb., 56035).Japan: Prov. Awaji, A. Yasuda, 111 (in Mo. Bot. Gard. Herb., 57027).

## 103. C. vinaceum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, very thin, closely adnate, drying between light pinkish cinnamon and vinaceous-buff, even, not shining, not cracked, the margin similar, thinning out; in section 75–100  $\mu$  thick, colored near the substratum, with the hyphae  $2\frac{1}{2}\,\mu$  in diameter, densely longitudinally interwoven and conglutinate, not incrusted, bearing a hymenium 25  $\mu$  thick; no gloeocystidia; basidia not protruding; spores white in spore collection, even, subglobose, 7–8  $\times$  6–7  $\mu$ .

Fructifications 5-10 mm. in diameter, near together and becoming irregularly confluent over areas up to 4 cm. long, 1-2 cm. wide.

Under side of decaying coniferous plank. Alabama and Louisiana. March.

The distinguishing characters of this species are occurrence on coniferous wood in closely adnate, vinaceous fructifications, which are somewhat colored next to the substratum and have large spores.

Specimens examined:

Alabama: Montgomery, R. P. Burke, 271 (in Mo. Bot. Gard.

Herb., 57156).

Louisiana: St. Martinville, A. B. Langlois, df, type.

## 104. C. fuscostratum Burt, n. sp.

Type: in N. Y. State Mus. Herb., Mo. Bot. Gard. Herb., and Burt Herb.

Fructifications broadly effused, thin, tender, forming a thin, fragile, cartridge-buff to pale smoke-gray hymenial pellicle on an arachnoid or fibrillose, wood-brown subiculum, the hymenium cracking into small polygonal masses about 1 mm. in diameter, the margin colored like the substance, fimbriate; in section 120–300  $\mu$  thick, wood-brown, with the hyphae pale brownish,  $2\frac{1}{2}\,\mu$  in diameter, nodose-septate, sometimes incrusted; no gloeocystidia; spores hyaline, even, flattened on one side,  $3-4\times 2\,\mu$ .

Fructifications 3-6 cm. long, 2-3 cm. wide.

On bark of decaying *Pinus Strobus* and other conifers. Canada to Maryland and westward to British Columbia. August to December. Uncommon.

The fructifications of C. fuscostratum are characterized by a hymenial layer as thin, fragile, and cracked as that of C. arachnoideum or of C. centrifugum and a supporting layer underneath as colored as that of C. subcontinuum. Compare C. ochroleucum Bres. and C, olivaceo-album.

Specimens examined:

Canada: J. Macoun, 15; St. Lawrence Valley, J. Macoun, 29.

New York: Albany, H. D. House, type (in N. Y. State Mus. Herb., Mo. Bot. Gard. Herb., 63750, and Burt Herb.), and H. D.

House & J. Rubinger (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 7766); Round Lake, C. H. Peck, (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 42930).

Pennsylvania: Freeland, C. R. Orton & G. E. Broadbent, comm. by L. O. Overholts, 5166 (in Mo. Bot. Gard. Herb., 56359).

Maryland: Takoma Park, C. L. Shear, 963.

Michigan: East Tawas, J. R. Weir, 317 (in Mo. Bot. Gard. Herb., 6961); New Richmond, C. H. Kauffman, 86 (in Mo. Bot. Gard. Herb., 54327).

Wisconsin: Star Lake, J. J. Neumann, comm. by H. von Schrenk (in Mo. Bot. Gard. Herb., 42734).

British Columbia: Kootenai Mountains near Salmo, J. R. Weir, 503, 511 (in Mo. Bot. Gard. Herb., 63722, 5900).

105. C. atrovirens Fries, Epicr. 562. 1838; Hym. Eur. 651. 1874; Berkeley, Outl. Brit. Fung. 274. 1860; Sacc. Syll. Fung. 6: 614. 1888; Massee, Linn. Soc. Bot. Jour. 27: 155. 1890; Bresadola, Ann. Myc. 1: 96. 1903; Maire, Brit. Myc. Soc. Trans. 3: 172. pl. 16. 1910; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 243. 1911; Rea, Brit. Basid. 677. 1922.

Thelephora atrovirens Fries, Elenchus Fung. 1: 202. 1828.— Lyomyces caerulescens Karsten, Finska, Vet.-Soc. Bidrag Natur och Folk 37: 154. 1882.—Hypochnus chalybaeus Schroeter, Krypt.-Fl. Schlesien 3: 416. 1888.

Fructifications irregularly effused, thin, floccose-fibrillose or arachnoid, greenish glaucous blue to deep bluish gray-green, even, not cracked, the margin thinning out, with hyphae interwoven; in section 150–250  $\mu$  thick, colored like the hymenium, composed of long, slender, interwoven, colored hyphae 2–3  $\mu$  in diameter, not nodose-septate, not incrusted; no gloeocystidia; spores colored like the fructification, even, subglobose, 3–4  $\times$  2½–3½  $\mu$ , borne 4 to a basidium.

Fructifications 1-4 cm. long, 1-2 cm. wide.

On under side of decaying bark and fallen branches. In Europe, and from New Brunswick to South Carolina and in Illinois. September to December. Infrequent.

C. atrovirens is conspicuous by its fructifications blue-green in all parts. It is intermediate between Corticium and Hypochnus, being included in the former on account of the even spores.

Specimens examined:

Exsiccati: Sydow, Myc. Germ., 1432.

Finland: Mustiala, P. A. Karsten, authentic specimen of Hypochnopsis caerulescens.

Germany: Brandenburg, P. Vogel, in Sydow, Myc. Germ., 1432.

Poland: Russian Poland, Eichler, comm. by G. Bresadola.

Great Britain: Coed Coch (in Berkeley Herb. of Kew Herb.).

New Brunswick: Campobello, W. G. Farlow.

Vermont: Middlebury, E. A. Burt, 2 gatherings.

Massachusetts: Beverly, C. W. Dodge & D. H. Linder, A (in Mo. Bot. Gard. Herb., 63451); Stony Brook, G. R. Lyman, 129.

New York: Cascadilla, A. J. Pieters, comm. by Cornell Univ. Herb., 5256; Ithaca, G. F. Atkinson, 8202; Karner, H. D. House, 14.205 and an unnumbered specimen (in Mo. Bot. Gard. Herb., 44727, 54394); Syracuse, L. M. Underwood, 44 (in N. Y. State Mus. Herb., and Mo. Bot. Gard. Herb., 56088).

North Carolina: Blowing Rock, G. F. Atkinson, 4301.

South Carolina: Gourdin, C. J. Humphrey, 2586 (in Mo. Bot. Gard. Herb., 43119).

Illinois: Hallidayboro, C. J. Humphrey, 2125 (in Mo. Bot. Gard. Herb., 22086).

106. C. caeruleum (Schrad.) Fries, Epicr. 562. 1838; Hym. Eur. 651. 1874; Berkeley, Outl. Brit. Fung. 274. 1860; Berk. & Curtis, Grevillea 1: 178. 1873; Sacc. Syll. Fung. 6: 614. 1888; Massee, Linn. Soc. Bot. Jour. 27: 151. 1890; Bourdot & Galzin, Soc. Myc. Fr. Bul. 27: 232. 1911; Wakefield, Brit. Myc. Soc. Trans. 4: 119. pl. 3, f. 26. 1913; Coker, Elisha Mitchell Scientif. Soc. Jour. 36: 169. pl. 33, f. 1. 1921; Rea, Brit. Basid. 673. 1922.

Thelephora caerulea Schrader in De Candolle, Fl. Fr. 2: 107. 1815; Persoon, Myc. Eur. 1: 147. 1822; Fries, Elench. Fung. 1: 202. 1828.—Auricularia phosphorea Sowerby, Eng. Fungi, pl. 350. 1802.—Thelephora Indigo Schweinitz, Naturforsch. Ges. Leipzig Schrift. 1: 107. 1822.

Fructifications somewhat round, broadly effused, adnate, rather thick, membranaceous, separable when moist, indigoblue to induline blue, even, somewhat velvety, the margin thin-

ning out, concolorous or whitish; in section 200–500  $\mu$  thick, thickening by becoming stratose, the outer stratum deep blue, the hyphae thick-walled, interwoven, nodose-septate, not incrusted,  $3-4\frac{1}{2}\mu$  in diameter; no gloeocystidia; spores even,  $6-10 \times 4\frac{1}{2}-5\mu$ .

Fructifications 3-10 cm. in diameter.

On under side of decaying limbs of *Quercus* and other frondose species. In Europe, southern United States, Illinois, and Japan. August to November. Probably in quantity where found.

 $\bar{C}$ . caeruleum is easily recognized by its deep blue color and occurrence on fallen oak limbs.

Specimens examined:

Exsiccati: Cavara, Fungi Longobardiae, 13; Cooke, Fungi Brit., 221, and ed. II, 5; Libert, Pl. Crypt. Ard., 22; Ravenel, Fungi Am., 451; Ravenel, Fungi Car. 3: 27; de Thümen, Myc. Univ., 1207; Westendorp, Crypt. Belge, 767.

Denmark: Skarup, E. Rostrup, in de Thümen, Myc. Univ., 1207.

Italy: Cavara, in Cavara, Fungi Longobardiae, 13.

Belgium: in Westendorp, Crypt. Belge, 767.

France: Libert, in Libert, Pl. Crypt. Ard., 22; Corrombles, F. Fautrey, comm. by Lloyd Herb.

England: Chichester, in Cooke, Fungi Brit., ed. II, 5.

South Carolina: H. W. Ravenel, in Ravenel, Fungi Car. 3: 27; Aiken, H. W. Ravenel, Fungi Am., 451.

Georgia: Atlanta, E. Bartholomew, 5679 (in Mo. Bot. Gard. Herb., 44218).

Florida: Sanford, C. L. Shear, 5204 (in Mo. Bot. Gard. Herb., 62164).

Alabama: Auburn, F. S. Earle (in Lloyd Herb., 3450, Burt Herb., and Mo. Bot. Gard. Herb., 4851), Earle & Baker, comm. by A.B. Seymour (in Mo. Bot. Gard. Herb., 16394); G. L. Peltier (in Mo. Bot. Gard. Herb., 4684), A. H. W. Povah, 906 (in Mo. Bot. Gard. Herb., 58692), and F. A. Wolf (in Mo. Bot. Gard. Herb., 43983); Montgomery County, R. P. Burke (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61562), and 14 (in Mo. Bot. Gard. Herb., 16983).

Illinois: Anna, C. J. Humphrey, 1356 (in Mo. Bot. Gard. Herb., 42932).

Arkansas: Womble, W. H. Long, 19769 (in Mo. Bot. Gard. Herb., 8961).

Japan: Sendai, A. Yasuda (in Mo. Bot. Gard. Herb., 58236).

#### EXTRA LIMITAL SPECIES

#### 107. C. paniculatum Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, thin, adnate, somewhat membranaceous, small pieces separable, pinkish cinnamon in the herbarium, even, not shining, not cracked, the margin narrow, thinning out, with hyphae interwoven; in section 200  $\mu$  thick, not colored, composed of loosely interwoven, hyaline hyphae 3  $\mu$  in diameter, not incrusted, not nodose-septate, and of irregularly arranged gloeocystidia or conducting organs up to 30–75  $\times$  3–6  $\mu$ , flexuous or irregular in form; paraphyses brownish, giving their color to the hymenium, paniculately branched, with the ultimate branches very slender, projecting beyond the basidia and forming the hymenial surface; basidia cylindric-clavate, 30–40  $\times$  4½–6  $\mu$ ; no spores found.

Fructifications 2 cm. long, 5 mm. wide, confluent longitudinally. On small, decaying, frondose limbs. Paraguay. August.

C. paniculatum is distinguished among the Corticiums which have gloeocystidia by its pinkish cinnamon color and hymenial surface composed of conspicuous, somewhat colored, bushybranched paraphyses.

Specimens examined:

Paraguay: Paraguari, *Malme*, 1081, type, comm. by L. Romell, 331.

# SPECIES TOO INCOMPLETELY DESCRIBED FOR LOCATION AMONG PRECEDING SPECIES

108. C. dendriticum P. Hennings, Hedwigia 41: Beiblatt, 102.
1902; Sacc. Syll. Fung. 17: 168. 1905; v. Höhnel & Litschauer,
K. Akad. Wiss. Wien Sitzungsber. 116: 742. 1907.

Type: in Berlin Herb.

"Carnoso-ceraceum, pallide carneum, dendroideo-ramosum vel radiato-effusum, margine sicco reflexo, albo-villosulo; hymenio ceraceo, pruinoso carneo, sicco rimoso, basidiis clavatis, 2–4-sterigmatibus, 20–28  $\times$  7–8  $\mu$ ; sporis subglobosis, subroseis, levibus, 4–5  $\mu$ .

"San Jose de Costa Rica auf Stämmen von Orangen.—H. Pittier.

"Der Pilz bildet fleischige, dendritisch verzweigte, fleischrothe Lager, derselbe soll eine Krankheit der Stämme verursachen. Mit. C. salicinum Fr. und C. sarcoides Fr. verwandt."

Von Höhnel and Litschauer, in their study of the type specimen of C. dendriticum, found the spores  $10-11\times 8~\mu$ , 4 sterigmata constantly, and the fructifications seated upon a lichen instead of directly on the trunk of Citrus aurantium.

#### EXCLUDED SPECIES

Corticium ferax Ell. & Ev. Am. Nat. 31: 339, 1897; Sacc. Syll. Fung. 14: 219. 1899.

Sections of the type specimen in Ellis Coll. in N. Y. Bot. Gard. Herb. show this to be a Hyphomycete. A specimen under this name collected on coniferous wood, Beaver Meadow, Hull, Quebec, was communicated by J. Macoun as the *Corticium ferax* Ell. & Ev. of Canadian Cryptogams, 246, Nat. Hist. Survey of Canada Herb.; this is *Peniophora glebulosa*.

#### SUPPLEMENT

Since the publication of the earlier parts, the following species have been received which were not included in those parts or require further notice.

#### **ALEURODISCUS**

See also account of species of *Aleurodiscus* by Lloyd, Myc. Writ. **6:** Myc. Notes 62: 926. f. 1666-1688. 1920; 65: 1066. f. 2009-2012. 1921.

Aleurodiscus cerussatus (Bres.) v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 807. pl. 4, f. 1. 1907; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 351. 1913.

Corticium cerussatum Bresadola, Fungi Trid. 2:37. pl. 144, f. 3.

1892; I. R. Accad. Agiati Atti III. 3:112. 1897; Sacc. Syll. Fung. 11:126. 1895.—*Kneiffia cerussata* Bresadola, Ann. Myc. 1:104. 1903.

Type: in Burt Herb., an authentic specimen which is probably a part of the type.

Fructifications effused, closely adnate, thin, waxy, white at first, becoming between pinkish buff and cream-buff in the herbarium, even, somewhat pruinose under a lens, cracking at right angles when old into masses about 3–4 to a mm., the margin similar, thinning out; in section 100–150  $\mu$  thick; not colored, composed of suberect, interwoven, densely crowded hyphae about 2  $\mu$  in diameter and of very numerous gloeocystidia; gloeocystidia flexuous, 40–60  $\times$  4–6  $\mu$ ; bottle-brush paraphyses form the hymenial surface; spores hyaline, even, 12–15  $\times$  7–8  $\mu$ .

Fructifications 1-7 cm. long, 2-10 mm. wide.

On old, weathered, coniferous wood. Europe, Manitoba, and Oregon. June to October.

C. cerusatus is distinguished from our other species of Aleurodiscus with the exception of A. succineus, by having both gloeocystidia and bottle-brush paraphyses and from the latter by being effused.

Specimens examined:

Italy: Trient, G. Bresadola, part of type probably.

Manitoba: Binscarth, G. R. Bisby, 1050 (in Mo. Bot. Gard. Herb., 59037); Winnipeg, G. R. Bisby, 65 (in Mo. Bot. Gard. Herb., 57899), and comm. by L. O. Overholts, 7027 (in Mo. Bot. Gard. Herb., 57475).

Oregon: Granite Pass, J. R. Weir, 8682 (in Mo. Bot. Gard. Herb., 36743).

A. disciformis (DC.) Patouillard, Soc. Myc. Fr. Bul. 10: 80. text f. 1894; v. Höhnel & Litschauer, K. Akad. Wiss. Wien Sitzungsber. 116: 798. pl. 1, f. 1. 1907; Bourdot & Galzin, Soc. Myc. Fr. Bul. 28: 350. 1913; Rea, Brit. Basid. 671. 1922.

Thelephora disciformis De Candolle, Fl. Fr. 6: 31. 1915; Fries, Syst. Myc. 1: 443. 1821.—Stereum disciforme (DC.) Fries, Epicr. 551. 1838; Hym. Eur. 642. 1874; Patouillard, Tab. Anal. Fung. 112. f. 250. 1884.—Peniophora disciformis (DC.) Cooke,

Grevillea 8: 20. pl. 122, f. 2. 1879; Sacc. Syll. Fung. 6: 642. 1888.

Fructifications effused, disciform, rather thick, pale olive-buff to cartridge-buff in the herbarium, pulverulent to velutinous, even, becoming somewhat cracked, the margin free, narrow, somewhat elevated, somewhat ochraceous on the under side; in section 150–800  $\mu$  thick, not colored, composed of erect, densely arranged hyphae 3–4  $\mu$  in diameter, with a great deal of crystalline matter intermixed; paraphyses  $4\frac{1}{2}$ –6  $\mu$  in diameter, cylindric, sometimes becoming irregularly swollen, sometimes somewhat moniliform toward the apex; spores hyaline, even, 15–20  $\times$  11–15  $\mu$ .

Fructifications  $\frac{1}{2}-2\frac{1}{2}$  cm. in diameter, or  $\frac{1}{2}-2\frac{1}{2}$  cm. long,  $\frac{1}{2}-1$  cm. wide.

On bark of Quercus. Europe, Mexico, and Africa. August to May.

A. disciformis is a species whose large fructifications resemble in aspect those of A. candidus although not as white as the latter and with paraphyses related in form to those of A. amorphus.

Specimens examined:

Austria: Vienna, V. Litschauer.

Italy: Trentino, G. Bresadola.

France: Aveyron, M. Galzin, 9503, comm. by H. Bourdot, 18550; locality not stated, Mougeot (in Farlow Herb.).

Mexico: locality not stated, A. Dampf (in Weir Herb.).

Africa: Union of South Africa, Stellenbosch, P. A. van der Bijl, 658 (in Mo. Bot. Gard. Herb., 59358).

# A. helveolus Bresadola, Mycologia 17: 71. 1925.

Type: in Weir Herb.

Fructifications erumpent, pulvinate to short-clavate, sessile, rugulose, waxy, somewhat gelatinous, Hay's brown, drying somewhat fuscous; hyphae hyaline, not incrusted,  $4\frac{1}{2}$ –6  $\mu$  in diameter; no conducting organs; basidia simple, large, 45–80  $\times$  6–8  $\mu$ , with 2–4 sterigmata; spores hyaline, even, 18–21  $\times$  6–9  $\mu$  according to Bresadola; hymenium surrounds the clubs on all sides.

Fructifications about 2 mm. high and 1 mm. in diameter when moistened.

On bark of dead Salix lasiandra. Washington. November.

The dried fructifications of A. helveolus have some resemblance in aspect to those of Stereum rufum but swell on softening and rise to a height of 2 mm. above the bark. The paraphyses were described by Bresadola as "paraphysibus irregularibus, undulatorestrictis, moniliformibus, laevibus, 3–6  $\mu$  crassis, apice interdum subcapitatis" but they do not show clearly in my preparation.

Specimens examined:

Washington: Spokane, alt. 576 m., J. R. Weir, 16312, type (in Weir Herb.).

A. macrodens Coker, Elisha Mitchell Scientif. Soc. Jour. 36: 155. pl. 15, upper figs., pl. 31, f. 7-9. 1921.

Type: part of type in Mo. Bot. Gard. Herb.

"Forming irregular, often somewhat elongated patches about 2 mm. to 2 cm. long with well-defined margins and with much the aspect of A. candidus; surface minutely pulverulent, pure white, or pale cream when old and weathered; entire thickness only about 150–190  $\mu$ , the structure in section much obscured by very small crystals and the densely branched paraphyses. Basidia entirely embedded, 12–15  $\mu$  thick, irregular and bent, with 4 long, stout sterigmata, which only reach the surface by their tips. Spores commonly rectangular in outline, the surface set with a few large, irregularly placed, bluntly pointed spines which are up to 4  $\mu$  long; body of spore  $11\frac{1}{2}-15 \times 18\frac{1}{2}-27 \mu$ ."

On bark of living trees of *Fraxinus* and *Salix*. New Hampshire to North Carolina. May to December. Probably common.

"In passing the plant would be taken for A. candidus, but when examined is seen to be much thinner with the closely pressed margin not showing a dark underside. The spores are remarkable and unlike any others in the genus."

Specimens examined:

New Hampshire: Chocorua, W. G. Farlow, 1.

New York: Alcove, C. L. Shear, 1302, 1305; East Galway, E. A. Burt; Poughkeepsie, W. R. Gerard, 294, comm. by N. Y. Bot. Gard. Herb.

North Carolina: Chapel Hill, W. C. Coker, 4734, type, comm. by Univ. North Carolina Herb. (in Mo. Bot. Gard. Herb., 57427).

A. subcruentatus (Berk. & Curtis) Burt, Mo. Bot. Gard. Ann. 7: 237. 1920; Zeller, Mycologia 14: 179. 1922.

Stereum subcruentatum Berk. & Curtis, Am. Acad. Arts & Sci. Proc. 4: 123. 1858; Sacc. Syll. Fung. 6: 567. 1888.

Type: in Farlow Herb.

Fructifications small, sometimes effuso-reflexed, with the reflexed portion up to 1-2 mm, broad but more frequently resupinate, somewhat discoid, with the margin free all around and slightly elevated—in one fructification grown out so as to be attached by the vertex; upper side of reflexed pileus whitish at the margin, avellaneous nearer the substratum, somewhat radiately rugose, mealy; hymenium even, white or becoming pinkish buff: pulverulent; in section 500-1000 \( \mu \) thick, not colored, composed of suberect, densely interwoven hyphae among a great amount of obscuring crystalline and mineral matter which is often in masses up to  $45 \times 15 \mu$ ; hyphae about  $2 \mu$  in diameter; hymenial portion up to 600 \(\alpha\) thick, composed of several layers, containing more or less numerous imbedded spores resembling the basidiospores; paraphyses simple, filiform, probably torulose, about 2-3 µ in diameter, basidiospores copious at surface of hymenium, hyaline, even, somewhat flattened on one side, 12- $18 \times 9 - 12 \, \text{u}$ .

Fructifications 2-15 mm, in diameter.

On bark of *Tsuga Sieboldii* in Japan and on bark of living trunks of *Picea sitchensis* and Douglas fir in California and Oregon. August and September.

A. subcruentatus has hymenial surface and spores suggestive of A. disciformis but is a very distinct species by having its fructifications effuso-reflexed when on the bark of standing trunks, by occurrence on conifers, thick and zonate hymenial portion, and presence of imbedded spores.

Specimens examined:

Oregon: Corvallis, S. M. Zeller, 1809 (in Mo. Bot. Gard. Herb., 56330).

California: Requa, W. H. Snell (Mo. Bot. Gard. Herb., 55860) and E. E. Hubert, comm. by J. R. Weir, 9946 (in Mo. Bot. Gard. Herb., 56229).

Japan: C. Wright, 265, type, Fungi U. S. Pac. Expl. Exp. (in

Farlow Herb.); Mt. Akayu, Prov. Echègo, A. Yasuda, 22 (in Mo. Bot. Gard. Herb., 55659).

A. succineus Bresadola, Mycologia 17: 71. 1925.

Type: in Weir Herb.

Fructifications small, flattened, becoming disk-shaped by slight elevation of the margin, mouse-gray, pruinose, with the margin thick, entire, becoming free, under side pale; in section 500  $\mu$  thick, composed of densely arranged, ascending, thin-walled, hyaline hyphae 3–5  $\mu$  in diameter and of numerous gloeocystidia; gloeocystidia flexuous, 75–100  $\times$  8–10  $\mu$ ; paraphyses cylindric, of bottle-brush form, very numerous in the surface of the hymenium; basidia with 4 sterigmata; spores hyaline, even, ellipsoidal, 10  $\times$  5  $\mu$ .

Fructifications 1-3 mm. long, 1-2 mm. wide.

On old weathered wood of Arbutus Menziesii. Oregon. September.

A. succineus is readily recognized by its discoid fructifications which have both gloeocystidia and bottle-brush paraphyses.

Specimens examined:

Oregon: Grants Pass, J. R. Weir, 8682, type (in Weir Herb.).

A. Zelleri Burt, n. sp.

Type: in Burt Herb.

Fructifications resupinate, gregarious, erumpent, pulvinate, convex, pinkish buff to tawny; in section about 600  $\mu$  thick, composed of a broad layer of erect, somewhat interwoven hyphae 3–3½  $\mu$  in diameter, not incrusted, bearing a hymenial layer; no cystidia; gloeocystidia flexuous, 30–40  $\times$  4  $\mu$ , confined to the hymenial layer; basidia protruding, with 4 sterigmata; spores hyaline, even, 6–9  $\times$  4–4½  $\mu$ , copious.

Fructifications  $\frac{1}{2}-1\frac{1}{2}$  mm. in diameter, about  $\frac{1}{2}$  mm. thick—10 on an area about 1 cm. square.

On small dead twigs of a frondose species—perhaps Alnus. Oregon. December.

A. Zelleri may be recognized by its small, tawny, convex fructifications, erumpent from lenticels in the bark and having somewhat the aspect of a Tubercularia.

Specimens examined:

Oregon: Corvallis, S. M. Zeller, 6800, type.

#### **CONIOPHORA**

Coniophora corrugis Burt, n. sp.

Type: in Burt Herb.

Fructifications broadly effused, coriaceous-membranaceous, loosely attached, separable when moist, between fawn color and salmon-pink to russet-vinaceous, even when dry, somewhat wrinkled when moist, cracking in drying, the margin whitish, byssoid; in section 300  $\mu$  thick, not colored, with a broad layer next to the substratum of slender, loosely interwoven, thickwalled, nodose-septate hyphae about  $3\frac{1}{2}-4~\mu$  in diameter, not incrusted, and with a very compact hymenial layer; no gloeocystidia nor cystidia; basidia with 4 sterigmata; spores even,  $6-10\times4-7~\mu$ , usually hyaline but when fully mature some at least are colored.

Fructifications 2-10 cm. long, 1-3 cm. wide.

On logs and dead limbs and on living trees of *Pinus ponderosa*, *Abies lasiocarpa*, *Picea Engelmannii*, *Juniperus*, and *Ribes*. In mountain forests. Wyoming to Colorado and British Columbia to Arizona. May to October. Common.

This species is most likely to be referred to Corticium, for it does not produce spores copiously and the few found in preparations may be full-sized and hyaline. It was 14 years after the type collection was received before it was demonstrated from a more mature specimen that the spores become colored finally. Several other collections with hyaline spores were received in the interval. C. corrugis may be recognized among our alpine species by its occurrence on the hosts stated, somewhat coriaceous, loosely attached, vinaceous fructifications, and large spores. The occurrence on living trees, as noted by Dr. Weir on Idaho specimens, is almost sufficient to identify this species when so found. C. corrugis seems related to C. polyporoidea.

Specimens examined:

Exsiccati: Baker, Pacific Slope Fungi, 3570, under the name Corticium corruge Burt.

Wyoming: Jackson Hole, E. B. Payson, 2369 (in Mo. Bot. Gard. Herb., 57369).

Colorado: Arapahoe region, B. M. Duggar (in Mo. Bot. Gard. Herb., 63771); Tolland, L. O. Overholts, 1801 (in Mo. Bot. Gard. Herb., 43785, 54873), and E. Bethel (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 61447).

Idaho: St. Joe National Forest, J. R. Weir (in Mo. Bot. Gard. Herb., 43759, 63761); Victor, E. B. Payson, 2353, 2362 (in Mo. Bot. Gard. Herb., 57358, 57362).

British Columbia: Sidney, J. Macoun, 83 (in Mo. Bot. Gard. Herb., 55354).

Washington: Mt. Paddo, W. N. Suksdorf, 732, type.

Oregon: Austin, J. R. Weir, 5242 (in Mo. Bot. Gard. Herb., 55944).

California: Mt. Shasta, E. B. Copeland, in Baker, Pacific Slope Fungi, 3570; Santa Barbara, W. H. Morse, comm. by C. J. Humphrey, 860 (in Mo. Bot. Gard. Herb., 19314).

Arizona: Mt. Humphrey, near Flagstaff, W. H. Long, 21323 (in Mo. Bot. Gard. Herb., 55130); Peak Agassiz, near Flagstaff, W. H. Long, 19489 (in Mo. Bot. Gard. Herb., 44737, 55129).

### C. flavomarginata Burt, n. sp.

Type: in Burt Herb.

Fructifications effused, thick, membranaceous, separable, when growing avellaneous, with the margin flavous, fading in the herbarium to pinkish buff with margin whitish, even or somewhat colliculose, velvety, the margin radiate-fimbriate; in section 500  $\mu$  thick, chamois-colored, becoming stratose, the hyphae suberect, densely arranged and interwoven, slightly colored, thinwalled, collapsing,  $3-3\frac{1}{2}$   $\mu$  in diameter, not incrusted, not nodose-septate; no cystidia nor gloeocystidia; spores slightly colored, even, cylindric,  $12-15 \times 4\frac{1}{2}-6$   $\mu$ .

Fructifications 1-3 cm. long, ½-3 cm. wide.

In crevices of the rough bark of large branches of Quercus Garryana. Washington. December and March.

The faded herbarium specimens of *C. flavomarginata* have aspect similar to those of *C. polyporoidea* but very different tissues and spores. The yellow margin of the thick, tan-colored fructifications composed of 3 strata, should make this species conspicuous in its region, and it is rather surprising that it has not been received except from Mr. Suksdorf.

Specimens examined:

Washington: Bingen, W. N. Suksdorf, 912, 913; W. Klickitat County, W. N. Suksdorf, 888, type, and 889.

### C. Sistotremoides (Schw.) Massee

Thelephora Sistotremoides Schweinitz, Naturforsch. Ges. Leipzig Schrift. 1: 109. 1822.—Corticium suffocatum Peck, N. Y. State Mus. Rept. 30: 48. 1879.

Type: under the name *Odontia Sistotremoides* of Curtis Herb. in Farlow Herb. and probably also in Berkeley Herb. at Kew and Schweinitz Herb.

I was misled as to C. Sistotremoides in my presentation of the species in Mo. Bot. Gard. Ann. 4: 249. 1917, by having to base the work on the descriptions formerly published. I have since found in Farlow Herb. a piece  $12 \times 6$  mm. of the authentic specimen from Schweinitz Herb. This specimen is in excellent preservation; a preparation from it wholly changes the concept of C. Sistotremoides, whose description should become:—

Fructifications effused, thin, membranaceous, not fleshy, somewhat separable, becoming sepia in the herbarium, even, not papillate; in section 200–300  $\mu$  thick, colored like the hymenium, composed of colored hyphae 4–4½  $\mu$  in diameter, incrusted, not nodose-septate, loosely arranged and interwoven, rather irregular in form; no cystidia present or not distinguishable from immature basidia; spores colored, even, 9–10  $\times$  6  $\mu$ .

Authentic specimen is on reddish brown coniferous bark.

The type specimen of *C. Sistrotremoides* is darker than that of *C. suffocata* but not specifically distinct in my opinion. The account and distribution published for the latter in my earlier work applies to *C. Sistotremoides*. The descriptive matter published there for *C. Sistotremoides* should be struck out.

### **CRATERELLUS**

Craterellus subundulatus Peck, N. Y. State Mus. Bul. 67: 27. 1903.

Thelephora subundulata Peck, Torr. Bot. Club Bul. 22: 492. 1895; Sacc. Syll. Fung. 14: 214. 1899.

Type: in N. Y. Bot. Gard. Herb.

Fructifications gregarious or cespitose; pileus thin, coriaceous-fleshy, depressed or subinfundibuliform, sometimes split on one side, slightly floccose-squamulose or fibrillose, grayish or grayish brown, becoming light drab in the herbarium, wavy or lobed on the margin, the lobes often overlapping; stem equal, solid, colored like the pileus; hymenium uneven or shallowly radiately venose, decurrent, drying light pinkish cinnamon; no setae nor cystidia; basidia with 4 sterigmata; spores hyaline, even, flattened on one side,  $6-9 \times 4\frac{1}{2}-6 \mu$ .

Fructifications when dried  $1\frac{1}{2}$ -2 cm. high; pileus 4-13 mm. in diameter; stem 8-14 mm. long,  $1-1\frac{1}{2}$  mm. thick.

On ground under trees of Fagus. New York and Delaware. July and August.

Peck noted this species as related to *C. sinuosus*, from which it differs in smaller size, solid and darker-colored stem, and slightly smaller spores. The fructifications are apparently plentiful when found, for some 30 fructifications of various sizes comprise each gathering.

Specimens examined:

New York: New York Botanical Garden, New York, *Peck & Earle*, 1064 (in N. Y. Bot. Gard. Herb., Mo. Bot. Gard. Herb., and Burt Herb.).

Delaware: Wilmington, A. Commons, 2718, type (in N. Y. Bot. Gard. Herb.).

## C. turbinatus Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Pileus solitary, stipitate, coriaceous-corky, cylindric-turbinate, solid, with the margin erect, lobed, thinner, and the disk depressed, drying snuff-brown to Prout's brown, glabrous, even; flesh drying pale Saccardo's umber, and with a fragrant, pronounced odor, and taste not noteworthy; lamellae decurrent, distant, narrow, about 1 mm. broad in the dried specimen, thin, about 2–4 mm. apart, not connected nor with venose interspaces, concolorous with the pileus, with colored conducting organs in the subhymenial tissue; basidia simple, with at least 2 sterigmata demonstrated; spores slightly colored, even, globose, 5–6  $\mu$  in diameter; stem not sharply differentiated from the pileus, solid, contracting abruptly below, glabrous.

Fructifications 10 cm. high; pileus 7 cm. high, 3-4 cm. in diameter, with lobes up to 3 cm. long; stem 3 cm. long.

On stump of Quercus. California. March.

I have seen of this species only a dried specimen which was collected by Lieutenant McWhorter at a military training camp and I am not sure that the species may not be transferred eventually to perhaps *Paxillus* on account of the thin lamellae, which are, however, very narrow and distant. The species is distinguished by its thick, solid, snuff-brown, glabrous fructifications drying with fragrant odor, by globose, colored spores, and by occurrence on an oak stump.

Specimens examined:

California: near Base Hospital, Camp Stewart, Palo Alto, F. P. McWhorter, type (in Mo. Bot. Gard. Herb., 57269).

Craterellus (?) Zelleri Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Pileus fleshy when growing, thin, tubaeform, drying Prout's brown, with the erect, spreading margin deeply lacerate—in some cases to the stem and rarely splitting the stem on one side nearly to the ground; stem short, perforate, hollow, even, glabrous, Prout's brown; hymenium drying chamois to Naples yellow, even or reticulately plicate and with the larger pores subdivided into smaller, shallow pits more completely covering the under surface of the pileus but present also, although less well-developed, in patches on the upper side; no gloeocystidia; basidia simple, with 6, or perhaps more, sterigmata; spores colored, even,  $8-9 \times 4\frac{1}{2}-6 \mu$ .

Fructifications up to 6 cm. high; pileus 3-4 cm. broad; stem 2 cm. long, 3 mm. thick.

On the ground in a dense forest. Oregon. March.

I have included this species in *Craterellus* because of the similarity of the subhymenial hyphae to the longitudinally arranged hyphae of the pileus and my inability to detect any evidence of an underlying hymenium. The aspect of the fungus is that of *Craterellus cornucopioides*. It is my opinion that this species will eventually be demonstrated to be a *Merulius* parasitic or saprophytic on the pilei of *Craterellus cornucopioides*. I know no *Merulius* to which this species is referable.

Specimens examined:

Oregon: Corvallis, S. M. Zeller, 2093, type (in Mo. Bot. Gard. Herb., 58770).

#### **CYPHELLA**

Cyphella alboviolascens (Alb. & Schw.) Karsten, Finska Vet.-Soc. Bidrag Natur och Folk 37: 133. 1882; 48: 400. 1889; Sacc. Syll. Fung. 6: 669. 1888; Bourdot & Galzin, Soc. Myc. Fr. Bul. 26: 225. 1910; Rea, Brit. Basid., 698. 1922; Pilat, Ann. Myc. 22: 211. 1924; Monogr. Cyphellacearum Czechoslov. 2: 45. pl. 1, f. 2. 1925.

Peziza alboviolascens Albertini & Schweinitz, Consp. Fung. 322. pl. 8, f. 4. 1805.—Cyphella Curreyi Berk. & Broome, Not. Brit. Fungi, 935, Ann. & Mag. Nat. Hist. III. 7: 379. 1861.

Fructifications gregarious or scattered, somewhat spherical at first, becoming flattened at the pore and somewhat hemispherical, white, densely villose, sessile or subsessile, soft throughout and easily sectioned, the margin inrolled; hairs white, rough, 6  $\mu$  in diameter, up to 120  $\mu$  long; hymenium concave, often violaceous; spores hyaline, even, flattened on one side, the convex side nearly subangular, 9–12  $\times$  6–9  $\mu$ .

Fructifications up to 1 mm. broad in American gatherings, up to  $\frac{1}{2}$  mm. high.

On dead twigs of *Syringa vulgaris* and *Sambucus*. Europe and Maine. July to October.

C. alboviolascens differs from C. Tiliae by softer fructifications, shorter, nearly 3-angled spores, and shorter hairs. C. villosa is closely related.

Specimens examined:

Exsiccati: Sydow, Myc. Germ., 353.

Germany: Brandenburg, P. Vogel, in Sydow, Myc. Germ., 353.

Czecho-Slovakia: A. Pilat.

Maine: Kittery Point, R. Thaxter (in Mo. Bot. Gard. Herb., 58742, and Burt Herb.), comm. by W. G. Farlow (in Mo. Bot. Gard. Herb., 55573).

# C. fasciculata (Schw.) Berk. & Curtis

Collections made on Alnus oregana extend the range of C. fas-

ciculata to Oregon. These specimens have the spores up to  $8-10 \times 5-6 \mu$ —twice the diameter of the spores of specimens of eastern United States—and somewhat larger basidia, but their other characters are so similar to those of eastern specimens that it now seems best to refer them to C. fasciculata.

These specimens are:

Oregon: Corvallis, F. D. Bailey (in Mo. Bot. Gard. Herb., 44144, 44199).

## C. galeata (Schum.) Fr.

To my description of this species in Mo. Bot. Gard. Ann. 1: 362. 1915, it should be added that the spores are tawny, rough to verrucose,  $7-9 \times 6-8 \mu$ , or subglobose,  $8-10 \mu$  in diameter, according to Bourdot & Galzin, Soc. Myc. Fr. Bul. 26: 227. 1910, and Rea, Brit. Basid., 704. 1922.

C. marginata McAlpine, Fung. Dis. Stone-fruit Trees in Australia, 120. f. 229-232. 1902; Sacc. Syll. Fung. 17: 192. 1905; Zeller, Mycologia 14: 179. 1922.

Fructifications gregarious, fleshy-gelatinous, sessile, globose, somewhat ochraceous, drying drab and hoary, the pore distinct when full grown but nearly closed by the inrolled margin; hairs curved, honey-yellow, even, up to  $120\times4~\mu$ ; basidia simple,  $40\text{--}45\times6\text{--}8~\mu$ , with 4 sterigmata; spores hyaline, even,  $10\text{--}12\times6\text{--}7~\mu$ .

Fructifications usually  $\frac{1}{2}$  mm. in diameter, reported up to 1 mm. in diameter.

On small "die back" twigs of peach, almond, and apple. Australia and Oregon. July.

The small, grayish drab fructifications were very numerous on the small twigs received. Up to 30 were counted on an area 1 mm square.

Specimens examined:

Oregon: Corvallis, S. M. Zeller, 1830, 1831 (in Mo. Bot. Gard. Herb., 56334, 56335).

C. muscicola Fries, Syst. Myc. 2: 202. 1823; Hym. Eur. 663.
1874; Patouillard, Tab. Anal. Fung. 19. f. 31. 1883; Sacc. Syll. Fung. 6: 682. 1888.

Phaeocyphella muscicola (Fr.) Rea, Brit. Basid., 704. 1922; Pilat, Monogr. Cyphellacearum Czechoslov. 2: 67. text f. 16. 1925.

Fructifications gregarious, sessile or subsessile, cup-shaped, thin, membranaceous, the margin slightly downy, at length somewhat flaring; hymenium concave, even, snuff-brown with the copious spores; spores colored, even, spherical,  $6-6\frac{1}{2}\mu$  in diameter, so copious that they conceal the basidia.

Fructifications up to 1 mm. in diameter in American specimens, equalling the diameter in height.

On mosses. West Indies. November.

I have seen no European specimens of this species but the single gathering from Grenada agrees well with the concept of the species as more definitely described by the recent European mycologists. The occurrence on mosses, ashy white, open cups which become slightly flaring at the margin, and brown hymenium and spores are distinctive characters.

Specimens examined:

Grenada: R. Thaxter, comm. by W. G. Farlow, 5.

C. patens A. L. Smith, Linn. Soc. Bot. Jour. 35: 10. pl. 1, f. 6-8. 1891: Sacc. Svll. Fung. 17: 192. 1905.

Type: in Brit. Mus. Herb. presumably.

"Sparsa, tubaeformis, dein elongata, fere ad basim fissa et expansa, margine superiore incurvata, circa 5 mm. longa, 2 mm. lata, extus flava tomentosa; hymenio brunneo, lamellis paucis angustis lamelliformis instructis; sporis globosis, minute asperulis,  $5 \mu$  diam., hyalinis.

"On bark of tree, Morne Niger Maron [Dominica]. Sept. 1892. No. 323.

"This species seems to form a transition between the forms with a rugulose hymenium such as *C. Malbranchei*, Pat., and genera with regular gills such as *Lentinus*; the incurving margin and the shape of the immature specimens have decided the placing it in *Cyphella*."

C. sessilis Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications gregarious, sessile, closely adnate, white, very thin, membranaceous-fleshy, applanate, even, ceraceous, the margin slightly elevated, narrow, white, fibrillose; in section 60  $\mu$  thick, not colored, with the hyphae ascending, thin-walled, 2-3  $\mu$  in diameter; no gloeocystidia; basidia simple,  $12 \times 4\frac{1}{2} \mu$ , with 4 sterigmata; spores becoming pale-colored, even,  $6-7 \times 3\frac{1}{2}-4 \mu$ .

Fructifications 200–400  $\mu$  in diameter.

On fallen palm leaves. Bermuda. January.

The small, circular fructifications are rather near together and numerous, 17 having been counted on an area 1 cm. square. They are adnate by the whole under surface, with the hymenium flat and bordered by the narrow, white, fibrillose margin. Most of the spores are hyaline; some, however, are somewhat colored. The aspect is that of a minute Discomycete.

Specimens examined:

Bermuda: H. H. Whetzel, Ajj, type, comm. by R. Thaxter (in Mo. Bot. Gard. Herb., 58708), and duplicate from H. H. Whetzel.

C. tela (B. & C.) Massee, Jour. Myc. 6: 179. pl. 7, f. 12, 13. 1891.

Peziza tela Berk. & Curtis, Grevillea 3: 156. 1875.—Tapesia tela (B. & C.) Sacc. Syll. Fung. 8: 373. 1889.—An Peziza Daedalea Schw.?

Type: in Farlow Herb. and Kew Herb., under the name Peziza tela.

"Gregarious on a dense white subiculum; cups minute, 150–180  $\mu$  diameter, subglobose; mouth at first small, becoming expanded, but the acute margin always remains more or less incurved; externally blackish brown, frosted with glistening crystals of oxalate of lime; hymenium concave, even, naked, blackish brown; basidia clavate, tetrasperous; spores subglobose or broadly pyriform, smooth, pale brown, 7 by 5  $\mu$ .

"On wood. Lower Carolina. (Type in Herb. Berk., Kew, No. 7724).

"The present species, owing to its dark color and gregarious habit, also being furnished with a dense, white, broadly effused,

superficial mycelium, suggests the genus Peziza when examined under a low power, but is a true Cyphella."

I have examined superficially the type of *Peziza tela* B. & C. in Farlow Herb. and the aspect is so similar to that of *Solenia poriae-formis* that Massee's statement about the spores of *P. tela* being colored should be confirmed. I was unable to make such examination of the spores. The type of *Peziza Daedalea* Schw. has the same aspect as *P. tela*.

### C. Thaxteri Burt, n. sp.

Type: in Burt Herb.

Fructifications very small, gregarious, stipitate, cup-shaped with the mouth open, drying between avellaneous and light pinkish cinnamon, merely farinose under a lens but really hairy when highly magnified, the margin inrolled when dry; hairs Isabella color, even, flexuous,  $25-30 \times 4-4\frac{1}{2} \mu$ ; hymenium Isabella color; basidia simple,  $16 \times 4-6 \mu$ ; spores ochraceous, even,  $7-8 \times 5 \mu$ ; stem central, cylindric, with surface like the pileus.

Fructifications about  $\frac{1}{4}$  mm. in diameter; stem about  $140 \mu$  long,  $60-80 \mu$  thick.

On bark. West Indies. November.

About 30 of the small, goblet-shaped fructifications are present on an area about ½ cm. long, ¼ cm. wide. The farinose surface of the exterior of the cups and stem is probably due to granular matter on the hairs, but no trace of such matter is found when the hairs are examined in permanent glycerine mounts by the compound microscope.

Specimens examined:

Grenada: Grand Etang, R. Thaxter, type, comm. by W. G. Farlow.

#### **HYPOCHNUS**

Hypochnus albus Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, small, circular, closely adnate, very thin, snow-white, even, velutinous under a lens with the large cystidia, not shining, the margin similar; in section 30–60  $\mu$  thick, not colored, composed of loosely interwoven, hyaline hyphae  $1\frac{1}{2}$ -2  $\mu$  in diameter, not nodose-septate, incrusted in the sub-

hymenium; no gloeocystidia; cystidia somewhat incrusted, 75–120  $\times$  9–15  $\mu$ , of greatest diameter at the base, usually seated on the incrusted zone, more rarely on the substratum; paraphyses delicate, branching in antler-shaped form; spores hyaline, globose,  $7\frac{1}{2}\mu$  in diameter, even at first, finally minutely echinulate, borne 4 to a basidium.

Fructifications 1-4 mm. in diameter, 3 present on an area  $12 \times 15$  mm.

On bark of a frondose species among mosses and lichens in a moist, virgin forest. Mexico. January.

The small, white fructifications, conspicuous cystidia, antler-shaped paraphyses, and echinulate spores form a unique group of characters distinguishing H. albus. But for the echinulate spores this species could have been placed in *Peniophora* next to P. phyllophila.

Specimens examined:

Mexico: Orizaba, Nuevo, W. A. & E. L. Murrill, 749a, type, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54654).

# H. epiphyllus (Schw.) Burt, n. comb.

Hydnum epiphyllum Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 163. 1832.—Hypochnus granulosus (Peck) Burt, Mo. Bot. Gard. Ann. 3: 218. text f. 9. 1916, where additional synonymy is given.

Type: in Farlow Herb. from Schweinitz Herb. and probably in Schweinitz Herb. and at Kew, under the name *Hydnum epiphyllum*.

In Curtis Herb. of Farlow Herb. there are specimens of this species under the name *Hydnum epiphyllum*, collected in Alabama, *Peters*, 1124, and also under the herbarium name, *Odontia grandinia*, collector *Peters*, 1116.

# H. filamentosus Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, small, closely adnate, very thin, hypochnoid-arachnoid, Mars-brown but this color completely soluble in dilute potassium hydrate solution; hymenium not continuous and showing many ends of fibrils under a lens, the margin thinning out;

in section up to 90  $\mu$  thick in some places but with much thinner connecting portions and mycelial strands in the same section, colored but wholly bleached by action of potassium hydrate solution, the hyphae incrusted, 4–5  $\mu$  in diameter, often together in rope-like strands up to 18  $\mu$  in diameter with crystalline matter on the outer surface of the strands; basidia 15  $\times$  5  $\mu$ , with 4 sterigmata, protruding from the covering matter of the strands, few observed; spores attached to the basidia are hyaline (after treatment of the sections with potassium hydrate solution), subglobose, rough, 3  $\mu$  in diameter; no cystidia.

Fructifications 1-4 mm. in diameter, somewhat confluent for a length of 2 cm.

On small fragments of blackened, decaying wood of a frondose species—apparently on the under side next to the ground. Cuba. March.

The specimen upon which the description is based is scantily developed; collections with normal continuous hymenium will probably eventually be made. The distinguishing characters seem to be Mars-brown color, complete bleaching by potassium hydrate solution, numerous rope-like strands, hyphae thinwalled, incrusted, collapsing, and small subglobose spores.

Specimens examined:

Cuba: locality not stated, C. G. Lloyd, 424, type (in Mo. Bot. Gard. Herb., 55174).

### H. fumosus Fr.

To the synonymy of this species in Mo. Bot. Gard. Ann. 3: 239. 1916, should be added *Odontia tenuis* Peck, N. Y. State Mus. Rept. 44: 134. 1891. Study of the type in N. Y. State Mus. Herb. shows the outer surface overrun with the intricate, branching, anastomosing, mycelial threads, and the spores white, minutely echinulate,  $4-4\frac{1}{2} \times 2\frac{1}{2} \mu$ —both characteristic of *H. fumosus*.

## H. pallidofulvus (Peck) Burt, n. comb.

Zygodesmus pallidofulvus Peck, N. Y. State Mus. Bul. 105: 30. 1906; Sacc. Syll. Fung. 22: 1358. 1913.—Hypochnus subferrugineus Burt, Mo. Bot. Gard. Ann. 3: 210. 1916.

Study of the type of *Zygodesmus pallidofulvus* in N. Y. State Mus. Herb. shows the species to be an *Hypochnus* specifically the same as *H. subferrugineus*, which therefore becomes a synonym.

H. Rhacodium Berk. & Curtis in herb. under Stereum, n. sp.

Type: in Mo. Bot. Gard. Herb., Farlow Herb., and Kew Herb. Fructifications effused, very thick, crust-like and brittle when dry and fuscous or dusky drab externally and throughout, colliculose, not cracked, the margin unknown; in section  $1\frac{1}{2}$ –3 mm. thick, fuscous, composed (1) of a layer  $\frac{1}{2}$ –2 mm. thick next to the substratum, fibrous and tow-like, composed of loosely interwoven, thick-walled, rigid hyphae up to 6  $\mu$  in diameter, not incrusted, rarely nodose-septate, and (2) of a crust-like hymenial portion, composed of 1 or 2 layers with hyphae erect, densely crowded, colored, 4–5  $\mu$  in diameter, not incrusted, not nodose-septate, bearing basidia; no gloeocystidia nor cystidia; basidia with at least 2 sterigmata demonstrated; spores concolorous with the hyphae, aculeate-tuberculate, somewhat angular, the body 6–7  $\mu$  in diameter.

Fructifications probably large—known from fragments up to 2 cm. long,  $\frac{1}{2} \text{ cm. wide}$ .

On under side of decaying logs of apparently a frondose species. Pennsylvania.

H. Rhacodium has the aspect of a thick, dark fuscous, effused Hypoxylon. The hyphae of the under layer are brittle when dry so that the hymenial crust is very likely to split away from the substratum through this brittle layer. The specimens in Kew and Farlow Herbaria, communicated by Michener through Curtis, consist of the hymenial crust. Michener's own specimen, now in the Mo. Bot. Gard. Herb., has the whole fructification to the woody substratum. This species is related to H. umbrinus.

Specimens examined:

Pennsylvania: E. Michener, type, No. 1435 to M. A. Curtis (in Mo. Bot. Gard. Herb., 5095, in Farlow Herb., and Kew Herb. as Curtis Herb., 4061, under the herbarium name Stereum Rhacodium).

H. subviolaceus Peck, N. Y. State Mus. Rept. 47: 151. 1894; Sacc. Syll. Fung. 11: 130. 1895.

Type: in N. Y. State Mus. Herb. and Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, violet-gray at first, becoming drab in the herbarium, even, velutinous, the margin whitish at first, fibrillose; in section 90  $\mu$  thick, colored, composed of suberect and interwoven, densely arranged, thin-walled hyphae 2  $\mu$  in diameter, some hyaline and many colored and bushy-branched; spores nearly hyaline, globose, rough or minutely aculeate,  $4-4\frac{1}{2}\mu$  in diameter.

Fructification  $2\frac{1}{2}$  cm. long,  $1\frac{1}{2}$  cm. wide, broken off at both ends and on one side.

On badly decayed coniferous wood. Canada. September.

The aspect of *H. subviolaceus* is not hypochnoid but rather that of a very thin *Coniophora*. The occurrence on decorticated coniferous wood, drab color, system of bushy-branched, colored tissue in addition to, and somewhat masking, the usual hyphae, and the small nearly hyaline spores should aid in recognition of this species.

Specimens examined:

Canada: J. Macoun, type (in N. Y. State Mus. Herb.), and comm. by N. Y. State Mus. Herb., T 34 (in Mo. Bot. Gard. Herb.).

H. umbrinus (Fr.) Quelet, Fl. Myc. 2. 1888.

The above combination has priority over that in Mo. Bot. Gard. Ann. 3: 213. 1916, according to Wakefield, Brit. Myc. Soc. Trans. 6: 132. 1919. I have not access to a copy of the Quelet.

Upon reexamination of the sections in my preparation from the type of *Thelephora arachnoidea* Berk. & Br., I think that this is a *Septobasidium* as stated by Bresadola, Ann. Myc. 14: 241. This species should therefore be struck out in my work where given as a synonym of *H. umbrinus*.

Caldesiella viridis (Alb. & Schw.) Pat. Essai Taxon. 120. 1900; Rea, Brit. Basid. 651. 1922; Bourdot & Galzin, Soc. Myc. Fr. Bul. 40: 128. 1924.—Odontia viridis (Alb. & Schw.) Quelet, Fl. Myc. 434. 1888; Bresadola, I. R. Accad. Agiati Atti III. 3: 97. 1897.—Hydnum viride (Alb. & Schw.) Fries, Syst. Myc. 1: 421. 1821; Hym. Eur. 614, 1874.

This species has hypochnoid texture, color deep grape-green at first, fading to Vetiver green in the herbarium, and minutely echinulate spores slightly colored, about  $3\frac{1}{2}-5 \times 3-3\frac{1}{2}\mu$ . I have seen American collections from Vermont, Missouri, and British Columbia. The fructifications are sometimes so even that they might be referred to *Hypochnus*.

#### **PENIOPHORA**

Peniophora populnea (Peck) Burt, n. comb.

Stereum populneum Peck, N. Y. State Mus. Rept. 47: 145. 1894.

Type: in N. Y. State Mus. Herb. and Burt Herb.

Fructifications effused, often confluent, adnate, rather thin, small pieces separable when moist, brown tinged with liver color when fresh, becoming between Natal brown and Mars brown in the herbarium, not shining, somewhat colliculose, contracting in drying and cracking into angular masses \( \frac{1}{2} - 1\frac{1}{2} \) mm. in diameter, the margin thin, radiate-dentate, pale, drving slightly free in some places; in section 250-300 \( \mu \) thick, colored, 2-layered, with a broad layer next to the substratum composed of longitudinally arranged, crowded and densely interwoven, nearly hyaline hyphae 2½-3 μ in diameter, and with an equal, colored hymenial layer composed of erect, densely crowded and interwoven, brownish hyphae and brownish paraphyses and cystidia; no gloeocystidia; cvstidia heavily incrusted, very large, up to  $60-100 \times 20-25 \,\mu$ , at the surface of the hymenium but not protruding; paraphyses hair-like, colored, slender, 1-1½ u in diameter, branching at or near the tips into 2 or 3 short branches; basidia cylindric, 70- $90 \times 3-4 \mu$ , probably simple and with 4 very short sterigmata; spores hyaline, even,  $12-15 \times 4 \mu$ .

Fructifications  $3\frac{1}{2}$  cm. long, 3 cm. wide.

On bark of decaying *Populus tremuloides*. New York. August.

P. populneum should be recognized by its occurrence on poplar logs, liver color externally and colored substance, cracked hymenium, very large cystidia, and long and slender basidia. The

layer of hyphae longitudinally arranged along the substratum and the very long and slender basidia have made me question whether this species is not an *Auricularia* but I have been unable to demonstrate transverse septation in any of the basidia.

Specimens examined:

New York: Ray Brook, Essex County, C. H. Peck, type (in N. Y. State Mus. Herb., under the name Stereum populneum, and in Burt Herb.).

#### **STEREUM**

Stereum aculeatum (B. & C.) Burt, n. comb.

Thelephora aculeata Berk. & Curtis, Grevillea 1: 149. 1873; Sacc. Syll. Fung. 6: 523. 1888; Burt, Mo. Bot. Gard. Ann. 7: 232. 1920.

I now refer to S. aculeatum a small specimen received since the publication of the part on Stereum. This specimen has the component fructifications central-stemmed, laterally confluent, and resembling in aspect S. pallidum, but differing from the latter by the presence of gloeocystidia and the absence of cystidia; the spores are hyaline, even,  $5 \times 3\frac{1}{2}-4 \mu$ .

Fructifications 4 cm. high,  $2\frac{1}{2}$  cm. wide.

On the ground. South Carolina and Missouri. June and August.

Specimens examined:

South Carolina: Santee Swamp, H. W. Ravenel, 764, type (Curtis Herb., 2009, in Kew Herb., and Farlow Herb.); Clemson College, P. H. Rolfs, 1835.

Missouri: locality not stated, *Dr. Emig*, comm. by J. R. Weir, 18820 (in Mo. Bot. Gard. Herb., 58744, and Burt Herb.).

S. atrorubrum Ell. & Ev. Acad. Nat. Sci. Philadelphia Proc. 1890: 219. 1890; Sacc. Syll. Fung. 9: 225. 1891.

Type: in N. Y. Bot. Gard. Herb., and a fragment in Burt Herb.

"Fan-shaped or reniform, 1-3 cm. broad and long, coriaceous, thin, narrowed behind into a sessile base, hollow at first (about the same color as S. complicatum) and tomentose-pubescent with a few narrow faint zones, but when mature of a dull dark red (about the color of the pileus of Pol. lucidus) with the surface

glabrous and densely radiate-rugose, margin lobed and crisped and in some specimens proliferous, young hymenium yellow, becoming when old brick color when moist, paler when dry. In the mature state the 3-5 concentric zones are more distinct and slightly elevated. The specimens roll up in drying and become hard and brittle."

We have but very few strictly sessile or reniform species of Stereum, although sessile specimens of common effuso-reflexed species were described as distinct species; more collections of S. atrorubrum are needed to clear up this important character in this case. The upper surface of the fragment seen by me is now dusky brown to bone-brown, glabrous, shining, strongly radiately rugose and shallowly concentrically sulcate; hymenium even, glabrous, avellaneous; in structure about 800  $\mu$  thick, composed of (1) an intermediate layer of longitudinal, densely arranged, thick-walled, rigid hyphae  $3-3\frac{1}{2}\mu$  in diameter, (2) bordered on the upper side by an opaque, brown layer 60  $\mu$  thick which gives the color to the pileus, and (3) curving on the lower side into a hymenial layer 300  $\mu$  thick; no cystidia, gloeocystidia, nor conspicuous conducting organs; spores up to  $7 \times 2-2\frac{1}{2}\mu$  present but may not belong for only 2 seen.

The date of the collection—May—and appearance of the hymenium suggest a specimen of the preceding season which has held over through the winter and may have had somewhat different characters when growing. The very dark-colored, strongly radiating rugose upper side of the pileus is noteworthy.

Specimens examined:

British Columbia: on old logs, J. Macoun, 86, type, a fragment examined.

S. radicans (Berk.) Burt, Mo. Bot. Gard. Ann. 7: 108. pl. 3, f. 16. 1920.

In a collection of this species from Porto Rico, in Mo. Bot. Gard. Herb., 7585, the spores have become slightly colored, showing that this species belongs in *Thelephora*. The species is really an intermediate between *Stereum* and *Thelephora*, having the dense, intermediate layer of *Stereum* and also vesicular gloeocystidia in the hymenial layer. The spores are still hyaline in 3 of the 4 gatherings which I have studied.

## S. Underwoodii Burt, n. sp.

An Stereum induratum Berkeley, Linn. Soc. Bot. Jour. 16: 44. 1877?

Type: in Burt Herb.

Fructifications corky, not hard nor indurated, adnate, resupinate and effused, sometimes narrowly reflexed, the reflexed surface drab in the herbarium where young, nearly black where oldest, somewhat concentrically sulcate, fibrillose, not shining, the margin entire; hymenium warm buff to honey-yellow in the herbarium, even, velutinous; in section  $\frac{1}{2}$ -2 mm. thick, colored warm buff to tawny olive throughout, stratose, composed of densely interwoven, colored, rigid hyphae  $1\frac{1}{2}$ - $2\frac{1}{2}$   $\mu$  in diameter, highly branched and with many branches of more or less antler-shaped form; no cystidia, gloeocystidia, conducting organs nor imbedded spores; spores hyaline, even,  $10 \times 5 \mu$  but may not belong, only 1 seen.

Fructifications effused over areas 6 mm.-5 cm. long, 6 mm.-2 cm. wide, the reflexed margin 2-3 mm. broad.

On bark of Xolisima. West Indies and Brazil. September and April.

This species has the antler-shaped branching of hyphae characteristic of Hypochnus pallescens, H. peniophoroides, Asterostromella dura, and Stereum duriusculum. The narrowly reflexed margin is well shown by the specimens from Jamaica and is important for location of S. Underwoodii in Stereum. The Brazilian specimen was received from Bresadola under the name Stereum induratum Berk.—a species known only from a single collection made by the Challenger Expedition in the East Indies and described as pileate, conchiform, 3 inches across, and very hard. S. Underwoodii is soft, not at all hard, and does not turn the edge of the razor in sectioning. I have not yet been able to study the type of S. induratum.

Specimens examined:

Jamaica: base of John Crow Peak, L. M. Underwood, 2432, type, comm. by N. Y. Bot. Gard. Herb.; Cinchona, L. M. Underwood, 3128, comm. by N. Y. Bot. Gard. Herb.

Brazil: Blumenau, Dr. Möller, comm. by Bresadola under the name of Stereum induratum.

#### THELEPHORA

Thelephora lutosa Schw. See Burt, Mo. Bot. Gard. Ann. 1: 216. 1914.

This rare species has been known only from the type collection from Salem, North Carolina. There is now an additional gathering by Dr. W. A. Murrill, 404, from Mountain Lake, Virginia, July 8-14, of which a specimen is in the Mo. Bot. Gard. Herb. The specimen grew in clay ground in mixed woods; a fragment of buried rotten wood is attached to the short, radicated base. This specimen does not necessitate any change in the description. In the dried fructification the soft, fine pubescence of the upper side, and cream color externally and within are distinctive characters. The older portion of the hymenium has assumed a light drab color with the spores, which are slightly colored, angular,  $4-6 \times 4-4\frac{1}{2} \mu$ .

#### TULASNELLA

Tulasnella calospora (Boud.) Juel, K. Svenska Vet.-Akad. Bihang till Handl. Afd. III. 23<sup>12</sup>: 23. 1897; Bresadola, Ann. Myc. 1: 114. 1903.

Prototremella calospora Boudier, Jour. de Bot. 10: 85. text f. 1-4. 1896.—An Tulasnella rosella Bourdot & Galzin, Soc. Myc. Fr. Bul. 39: 263. 1924?

Fructifications effused, very thin, waxy, whitish in the herbarium, somewhat perforate, the margin thinning out; in section 100–150  $\mu$  thick, with the hyphae about 3  $\mu$  in diameter, thinwalled; spores hyaline, even, fusiform, flexuous,  $20-27 \times 3-3\frac{1}{2} \mu$ , often with a lateral branch.

Covering as a cluster of small fructifications the terminal portions of dead mosses on an area 2 cm. long, about 1 cm. wide.

On wood in Europe, on dead mosses in Maine.

T. calospora has fructifications rather more membranaceous than those of our other species, and longer spores, which are noteworthy by having frequently a branch stand out at right angles from the body of the spore. I figured such a branched spore in Mo. Bot. Gard. Ann. 6: 258. text f. 3. 1919.

Specimens examined:

Maine: Kittery Point, R. Thaxter (in Mo. Bot. Gard. Herb., 57477).

#### **VELUTICEPS**

Veluticeps fusca Humphrey & Long, n. sp.

Type: in Humphrey Herb. and Mo. Bot. Gard. Herb.

Fructifications coriaceous-corky, resupinate, effuso-reflexed, or conchiform, laterally confluent, with the reflexed part somewhat concentrically sulcate, tomentose, at first nearly auburn or tawny, finally becoming dusky drab and weathering hoary, the margin clay-colored when young, entire, becoming somewhat crisped; hymenium plane, avellaneous, velutinous, thickly studded with protruding fascicles of colored hyphae which have the appearance of teeth of a Hydnum when little magnified; in section 1-3 mm. thick, wood-brown, composed of densely arranged. suberect and interwoven, rigid, colored hyphae 3-41/2 \mu in diameter, not incrusted, not nodose-septate; hyphal fascicles 12-25 μ in diameter, protruding through and beyond the hymenium up to 90-150 μ and composed of flexuous, parallel, colored hyphae 3½-7 μ in diameter; basidia simple, with 4 slender, conspicuous sterigmata up to 6 µ long; spores white, even, usually unequilateral,  $9-10 \times 3\frac{1}{2}-4 \mu$ .

Confluent over areas up to 12 cm. long and 2-3 cm. wide, the reflexed margin 6-12 mm. broad.

On decorticated, decaying logs of *Pinus ponderosa*. Washington, Arizona, and New Mexico. October.

It is probable that V. fusca occurs more frequently than its few, widely separated, recorded stations indicate, for gatherings are likely to be referred by collectors to Hydnum on account of the superficial resemblance of the hymenial fascicles to teeth of Hydnum. The fructifications are large and conspicuous, somewhat resembling in aspect those of Stereum sulcatum but quite distinct by the hymenial fascicles.

Specimens examined:

Washington: Spokane, J. R. Weir, 611 (in Mo. Bot. Gard. Herb., 36749).

Arizona: Fort Valley Experiment Station, near Flagstaff, W. H. Long, 19688, type (in Mo. Bot. Gard. Herb., 20084).

New Mexico: Gila National Forest, near Pinos Altos, G. G. Hedg-cock & W. H. Long, 9851, comm. by C. J. Humphrey, 2572 (in Mo. Bot. Gard. Herb., 11200).

# AURICULARIACEAE SEPTOBASIDIUM

Septobasidium mexicanum Sydow, Ann. Myc. 18: 154. 1920; Sacc. Syll. Fung. 23: 567. 1925.

"Omnino resupinatum, matrici arcte adhaerens, tenuissimum, centro circiter  $\frac{1}{2}$ – $\frac{3}{4}$  mm. crassum, ca. 1–3 cm. longum, 1–2 cm. latum, ferrugineum, centro dein cinereo-ferrugineum, ad ambitum anguste sed distincte albido-cinereo fimbriatum, leve, haud rimosum; contextus ex hyphis flavo-brunneis crasse tunicatis 3–4  $\mu$  crassis sparse ramosis remote septatis compositus; basidia non visa.

"Hab. ad ramos vivos Cupressi spec., Mexico, 1918, leg. Reiche no. 46."

S. pedicellatum Patouillard, Jour. de Bot. 6: 61. text f. 1892; Burt, Mo. Bot. Gard. Ann. 3: 323. 1916.

Type: in Museum of Paris.

Since my account of this species I have studied specimens of both the Cuban collections distributed by C. Wright under the name *Thelephora pedicellata* and find that the collection, C. Wright, 798, distributed in Wright, 'Fungi Cubenses Wrightiani' is in condition to afford the structural details figured by Patouillard and therefore must be the type distribution of his species.

The general description of this species, which could not be given before, is:

Fructifications resupinate, dry, avellaneous, pulverulent, occurring in small, interrupted patches, each about 2–3 mm. in diameter; in structure 500–600  $\mu$  thick, colored, stratose, composed of 2 strata, each consisting of a hymenial crust supported on pillars or pedicels about 15  $\mu$  in diameter, with their component hyphae about 3  $\mu$  in diameter; probasidia borne at the surface of the hymenial layer.

On living bushes among, and on, mosses and lichens. Cuba.

S. pinicola Snell, Mycologia 14: 58. pl. 11-13. 1922; Overholts, Mycologia 16: 233. 1924.

Type: in Snell Herb., Mo. Bot. Gard. Herb., and Forest Path. Herb.

"Fructification resupinate, effused, coriaceous, in general circular in shape, more or less concentrically sulcate, separable from substratum, roughly tomentose to strigose, army-brown to Natal-brown when dry, the margin light drab to cinnamon-drab. strigose; in structure lacunar, spongy, 1-1.8 mm. thick, individual hyphae under the microscope clay-color to tawny olive, thickwalled, even, 3-3.5 \(\mu\) in diameter, loosely interwoven so as to form a spongy structure with locules, branching to form a lighter colored hymenium about 80-110 µ thick; probasidia terminal or lateral, hyaline, pyriform to subglobose,  $10-15 \times 15-17 \mu$ , throughout hymenium: spore-bearing organs straight, hyaline,  $54-66 \times 6-7 \mu$ , 3-septate, growing from probasidia and projecting above hymenium; spores hyaline, simple, curved, 14-17.5  $\times$  3-3.5  $\mu$ , borne singly from each of 3 cells of spore-bearing organ, acropetally as far as observed.

"Fructification 3-60 mm. but more commonly 10-35 mm. in diameter, 1-1.8 mm. thick."

On bark of living Pinus Strobus in New England, New York, and Pennsylvania, and probably co-extensive with the habitat of this host; also on Pinus monticola in Idaho. Found sporulating after prolonged moist and rainy period in August.

S. Spongia (Berk. & Curtis) Patouillard, Soc. Myc. Fr. Bul. 16: 1900; Burt, Mo. Bot. Gard. Ann. 3: 339. text f. 11.

From several collections of this species made by Dr. J. A. Stevenson in Porto Rico and San Domingo, additional characters have been secured for completion of the description.

Fructifications on leaves and stems of Citrus decumana and C. sinensis dry, warm sepia to Benzo-brown; probasidia at the hymenial surface of a few filaments are hyaline, globose, 9 µ in diameter; spore-bearing organs straight, cylindric-clavate; spores simple, hyaline, curved, 9-10  $\times$  3-4  $\mu$ , observed on the outer cells of the organs.

Sterile fructifications have been received from Dr. A. T. Speare, collected on Citrus, at Okeechobee, Florida.

### EXOTIC SPECIES

S. album Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications resupinate, effused, adnate, thick, fleshy, white, somewhat colliculose, pulverulent, contracting in drying and forming a few large fissures at 2–3 mm. apart, the margin somewhat tomentose; in structure 600–700  $\mu$  thick, not colored, composed of densely interwoven and ascending, even-walled, hyaline hyphae 3–4  $\mu$  in diameter, occasionally nodose-septate, not incrusted; no probasidia seen; spore-bearing organs straight, 3-septate, 75  $\times$  6  $\mu$ , confined to the outer 150  $\mu$  of the hymenium, only rarely reaching the surface and protruding; spores simple hyaline, even, 10–12–15  $\times$  7–9  $\mu$ , borne singly on the outer 3 cells of the spore-bearing organs so far as observed; surface of the hymenium composed of slender, hyaline, matted and coiled paraphyses or hyphal branches 2  $\mu$  in diameter.

Fructifications 1-3 cm. long, 1-1½ cm. wide.

On bark of dead, fallen branches of *Nothofagus*. New Zealand. December.

S. album somewhat resembles Corticium portentosum and is exceptional, if there is no error in the collector's data, by its occurrence on dead, fallen branches. The hymenial surface of coiled paraphyses, absence of probasidia, and hyphae extending from all parts of the substratum into the fructification without consolidation into supporting pillars are additional characters for recognition of the species.

Specimens examined:

New Zealand: Queenstown, Otago, G. H. Cunningham, 542, type, comm. by J. R. Weir (in Mo. Bot. Gard. Herb., 59315).

## S. cinnamomeum Burt, n. sp.

Type: in Farlow Herb. and Mo. Bot. Gard. Herb.

Fructifications resupinate, effused, dry, hypochnoid, small pieces separable when moist, Brussels brown, somewhat colliculose, somewhat velutinous, the margin concolorous, with surface irregular, somewhat raduloid; in section 210  $\mu$  thick, colored, stratose, consisting of 2 strata, each composed of suberect, loosely interwoven, rigid hyphae 3  $\mu$  in diameter, colored

like the hymenium, not incrusted, not nodose-septate; probasidia spherical,  $6 \mu$  in diameter; spore-bearing organs numerous, cylindric,  $30 \times 6 \mu$ ; spores simple, hyaline, curved,  $13 \times 3\frac{1}{2} \mu$ .

Fructifications probably large, for the one seen covered an area 7½ cm. long, 5 cm. wide.

On moss-covered bark of an apparently frondose species. Chile. December.

Distinguished by bright Brussels brown color, stratose structure consisting of 2 strata in the type, and absence of supporting pillars for the hymenial crust. The hyphae arise uniformly into the fructification from all points in the substratum.

Specimens examined:

Chile: Corral, R. Thaxter, b, type (in Farlow Herb., and Mo. Bot. Gard. Herb., 57896).

## S. spiniferum Burt, n. sp.

Type: in Farlow Herb. and Mo. Bot. Gard. Herb.

Fructifications resupinate, effused, adnate, coriaceous, blackish brown (3) in the herbarium, not shining, surface somewhat veined and with the veins extended into occasional, cylindric teeth or spines 2-5 mm. long, 2/5 mm. in diameter, extending obliquely from the veins and the hymenial surface in the marginal region, the margin fimbriate; in section 400  $\mu$  thick, colored, composed of loosely interwoven, rigid hyphae  $4\frac{1}{2}\mu$  in diameter, concolorous with the fructification; probasidia 9-12  $\mu$  in diameter; no spores nor spore-bearing organs seen.

Fructification 9 cm. long, surrounding a living hardwood branch 12 mm. in diameter.

On living, frondose branches. Chile. November.

The veined hymenium of S. spiniferum locates this species in the group with S. retiforme. The extension of the veins in the form of large hydnoid teeth is a unique character, if constantly present in future collections.

Specimens examined:

Chile: San Pedro, Concepcion, R. Thaxter, a, type (in Farlow Herb., and Mo. Bot. Gard. Herb., 57895).

### TREMELLACEAE

#### **EICHLERIELLA**

Eichleriella mexicana Burt, n. sp.

Type: in Mo. Bot. Gard. Herb. and N. Y. Bot. Gard. Herb.

Fructifications coriaceous, separable, effuso-reflexed, with the reflexed portion narrow, snuff-brown, and concentrically sulcate on the upper side, fibrillose, the margin entire; hymenium light vinaceous-cinnamon in the herbarium, pruinose, even; in section  $600~\mu$  thick, (1) with the layer at surface of pileus and next to substratum up to  $100~\mu$  thick and having its hyphae Saccardo's umber, loosely interwoven, thick-walled,  $3-4~\mu$  in diameter, and (2) with a broad intermediate layer composed of densely interwoven, hyaline hyphae  $4~\mu$  in diameter which passes into (3) the hymenial layer composed of basidia and slender branched paraphyses bearing granules; basidia immersed about  $30~\mu$  below the surface of the hymenium, longitudinally septate,  $16-21~\times~10-11~\mu$ ; spores simple, hyaline, even,  $12~\times~4-5~\mu$ .

Fructification resupinate over an area 4 cm. long,  $1\frac{1}{2}$  cm. wide, and broken off at both ends; the reflexed portion 2 mm. broad.

On bark of a decaying, frondose limb. Mexico. December. *E. mexicana* is related to *E. alliciens* but is thicker, browner above, with branched paraphyses bearing granules, and with larger spores.

Specimens examined:

Mexico: Guernavaca, W. A. & E. L. Murrill, 399, type (in Mo. Bot. Gard. Herb., 54547, and N. Y. Bot. Gard. Herb.).

#### **SEBACINA**

Sebacina (?) Cokeri Burt, n. sp.

Sebacina sp. Coker, Elisha Mitchell Scientif. Soc. Jour. 35: 157. pl. 47, 61, f. 1-5. 1920.

Type: in Univ. of North Carolina Herb. and Mo. Bot. Gard. Herb.

"Forming low, crowded and anastomosing, nodulated masses and pustules looking very like a Myxomycete; patches 9 cm. or more long and up to 1.5 cm. wide in our collection (probably quite indefinite as to size and form of area covered); height only up to

1 or 1.5 mm.; color a pallid creamy yellow or dusky cream; surface glabrous, shining unless getting rather dry. Texture succulent but not gelatinous in the usual sense, but firmly waxy. Fibers of the flesh slender and regular, about  $1.5-2~\mu$  thick, sparingly branched.

"Spores oval, flattened on one side, yellowish under microscope, very variable in size,  $6.3-9 \times 7.7-12.2 \,\mu$ , sprouting into threads by one or two germ tubes, which may arise at any point. Basidia oval,  $13.7-14.4 \times 16.3 \,\mu$ , irregularly four-celled, collapsing soon after formation of spores. Sterigmata much thickened upward, some very long and slender. Paraphyses slender, densely packed, curved over, and mostly branched a little at the ends, the branches crooked and rhizoid-like and more slender and set with very minute crystals. Much larger, roughly globular or angular crystals with slender, spine-like, hyaline projections also occur rather abundantly through the hymenium; they are mostly about  $7-9 \,\mu$  thick.

"This species is markedly distinct from all others we have seen. The peculiar color, pustulate, anastomosing form and plump spores and large crystals separate it easily from our other Sebacinas. The projections on the crystals do not seem to be of the same nature and after drying reappear very obscurely if at all. They may be the stubs of hyphae that took part in the formation of the crystals. So thickly interwoven are the tips of the paraphyses and so dense the little crystals that there is formed a distinct and darker crust over the surface."

The thickest portion of the fructification has dried Dresden brown.

Specimens examined:

North Carolina: Chapel Hill, on under side of old, hard heart of an oak branch, February, W. C. Coker, 4116, type (in Mo. Bot. Gard. Herb., 56719).

# S. fibrillosa Burt, n. sp.

Type: in Mo. Bot. Gard. Herb. and N. Y. Bot. Gard. Herb.

Fructifications effused, incrusting, adnate, rather thin, fibrillose-hypochnoid, drying whitish, somewhat velutinous, surface irregular and conforming to the elevations and depressions of the

surface upon which growing, the margin somewhat fimbriate; in section 200–400  $\mu$  thick, not colored, composed of densely interwoven, hyaline hyphae about  $2\frac{1}{2}\mu$  in diameter, with the wall gelatinously modified, much foreign matter present; cystidia not incrusted, cylindric, obtuse,  $3\frac{1}{2}-7\mu$  in diameter, protruding up to 30  $\mu$ ; basidia longitudinally septate, pyriform, 15  $\times$  9  $\mu$ , present in the surface of the hymenium; spores simple, hyaline, curved,  $7-8\times3\frac{1}{2}-4\mu$ , copious.

Fructification 3 cm. long, 2 cm. wide.

Running over wood humus on the forest floor at 7000 feet altitude. Mexico. December.

S. fibrillosa is a small, whitish, incrusting species running over the irregular surface of wood humus. Its distinguishing character is the presence of cystidia, which are conspicuous and as distinct as in a *Peniophora*, and locate this species in the subgenus *Heterochaetella* of *Sebacina*.

Specimens examined:

Mexico: Tepeite River region, near Guernavaca, W. A. & E. L. Murrill, 515, type (in N. Y. Bot. Gard. Herb., and Mo. Bot. Gard. Herb., 54514).

## S. lactescens Burt, n. sp.

Type: in Mo. Bot. Gard. Herb. and Farlow Herb.

Fructifications effused, rather thick when moist, thin when dry, gelatinous, separable, loosely attached, drying between drab and wood-brown, even, the margin thinning out; in section 1000  $\mu$  thick, not colored, composed of densely arranged, ascending and interwoven hyphae with walls so completely modified gelatinously that only the protoplasmic contents of the lumen can be followed; gloeocystidia somewhat colored, clavate, 54  $\times$  5–7½  $\mu$ , abundant in the hymenium; basidia longitudinally cruciately septate, 15  $\times$  12  $\mu$ , immersed about 25–35  $\mu$  below the surface of the hymenium; spores hyaline, even, curved, 12  $\times$  6  $\mu$ .

Fructifications 2 cm. long,  $\frac{1}{2}$ -1 cm. wide.

Longitudinally confluent on the under side of a frondose limb. West Indies.

S. lactescens may be recognized by its wood-brown color when dry, gelatinous consistency, and numerous and conspicuous,

slightly colored gloeocystidia. The latter locate this species in the subgenus *Bourdotia* of *Sebacina*.

Specimens examined:

Grenada: Grant Etang, R. Thaxter, comm. by W. G. Farlow, 153, type (in Mo. Bot. Gard. Herb., 55236).

- S. plumbescens Burt, Mo. Bot. Gard. Ann. 3: 241. 1916.
- S. plumbea Burt, Mo. Bot. Gard. Ann. 2: 765. text f. 6, pl. 27, 7. 20. 1915, but not of Bresadola & Torrend, Broteria 11: 87.
- f. 8. 1913.—S. Burti Trotter in Sacc. Syll. Fung. 23: 573. 1925.

### S. murina Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, very thin, pallid mouse-gray and somewhat pulverulent when dry, even, the margin similar; in section 30  $\mu$  thick, not colored, composed chiefly of longitudinally septate basidia starting almost directly from the substratum, 15  $\times$  8  $\mu$ , and of immersed, white, incrusted masses up to 25  $\times$  7  $\mu$  as seen in lactic acid preparations, densely covered with spiculose granules which clothe a short, cylindric, flexuous, hyphal axis for each mass; spores simple, hyaline, even,  $9\times 6~\mu$ .

Fructifications 5-6 cm. long,  $1\frac{1}{2}$ -2 cm. wide.

On decorticated, weathered, badly decayed wood on mountain side at altitude 800-1500 feet. Mexico. January.

S. murina is noteworthy by the small, erect, cylindric, incrusted, white masses between its basidia. These masses are evidently homologous with the paraphyses of S. calcea but differ from the latter by being unbranched, as shown when their spiculose, incrusting matter is cleared away by potassium hydrate solution; the central axis of each mass then becomes visible as a cylindric, flexuous rod somewhat olivaceous in color in preparations stained with eosin and very similar in appearance then to the organs termed gloeocystidia by Bourdot & Galzin in the subgenus Bourdotia of Sebacina.

Specimens examined:

Mexico: Motzorongo, near Cordoba, W. A. & E. L. Murrill, 986, type, comm. by N. Y. Bot. Gard. Herb. (in Mo. Bot. Gard. Herb., 54609).

S. polyschista Berk. & Curtis, n. sp., in herb. under *Corticium*. Type: in Farlow Herb. and probably in Kew Herb.

Fructifications effused, rather thin, loosely attached to the substratum, separable, fleshy, avellaneous in the herbarium, even, cracking in drying and showing through the cracks the whitish, fibrous subiculum, the margin thinning out, whitish, arachnoid; in section 400–500  $\mu$  thick, slightly colored, with the hyphae near the substratum loosely interwoven, thick-walled,  $4\frac{1}{2}$ –6  $\mu$  in diameter, not nodose-septate, not incrusted, and with the hymenial layer 200  $\mu$  thick, composed of densely interwoven hyphae 3  $\mu$  in diameter; no cystidia; basidia cylindric, apparently longitudinally septate, at the surface of the hymenium; spores hyaline, even, curved,  $10 \times 4\frac{1}{2}\mu$ .

On under side of limb of dead Pyrus Malus. South Carolina. July.

This species should be recognized by the avellaneous color of its fructifications which shrink greatly and crack in drying. It is related to S. adusta.

Specimens examined:

South Carolina: Society Hill, M. A. Curtis, 4950, type (in Farlow Herb.).

S. Sheari Burt, Mo. Bot. Gard. Ann. 2: 758. text f. 2. 1915.

This species was transferred to the genus *Heterochaete*, in Mo. Bot. Gard. Ann. 8: 377. 1921, under the name *Heterochaete Sheari* Burt.

### EXOTIC SPECIES

S. africana Burt, n. sp.

Type: in Mo. Bot. Gard. Herb.

Fructifications effused, closely adnate, thin, fleshy-gelatinous, drying cartridge-buff, contracting in drying and cracking, even, not shining, the margin not present; in section 240  $\mu$  thick, not colored, composed of suberect, densely arranged hyphae with walls gelatinously modified, somewhat granule-incrusted; gloeocystidia not colored, flexuous,  $75 \times 4-6 \mu$ , confined to the hymenial region between the basidia; basidia pyriform, at the surface of the hymenium; spores simple, hyaline, curved,  $6-7\frac{1}{2}\times 3 \mu$ .

Fructifications probably large, for specimen received is 9 cm. long, about 1 cm. wide, and broken off on all sides.

On decorticated, rotten, frondose log. South Africa. January.

S. africana resembles in aspect Corticium ochraceum but is a Sebacina in structure. It is further distinguished by its buff color, sparingly granule-incrusted, gelatinous-walled hyphae, small spores, and colorless, flexuous gloeocystidia which are in all respects like those present in some species of Corticium and Peniophora. The gloeocystidia locate S. africana in the subgenus Bourdotia of Sebacina.

Specimens examined:

South Africa: Knyna, Cape Colony, P. A. van der Bijl, 1342, type (in Mo. Bot. Gard. Herb., 63405).

#### TREMELLODENDRON

Tremellodendron simplex Burt, Mo. Bot. Gard. Ann. 2: 742. pl. 26, f. 5. 1915.

Another collection of this species, affording a more accurate description, consists of 2 infundibuliform fructifications with black, rugose, compressed stems; the pilei are olive-buff, even, glabrous; hymenium inferior, testaceous, with the margin olive-ocher.

Fructifications 3 cm. high; stem 2 cm. long,  $1\frac{1}{2}$  mm. in diameter; pileus 1 cm. in diameter, about 1 cm. long.

This gathering was made at El Yunque, Cuba, in March, 1903, by Underwood & Earle, 1087A, and is now in N. Y. Bot. Gard. Herb.

T. tenax (Schw.) Burt, Mo. Bot. Gard. Ann. 7: 67. pl. 11, f. 105, 106. 1922.

Clavaria tenax Schweinitz, Am. Phil. Soc. Trans. N. S. 4: 182. 1832.—Merisma tenax (Schw.) Léveillé, Ann. Sci. Nat. Bot. III. 5: 157. 1846.—Pterula tenax (Schw.) Sacc. Syll. Fung. 6: 742. 1888.—Tremellodendron Hibbardi Lloyd, Myc. Writ. 6. Myc. Notes 65: 1049. pl. 179, f. 1947. 1921.

Type: in Schweinitz Herb. and a fragment in Farlow Herb.

Fructifications fascicled, with substance very tough, at length somewhat horn-like, soon ramose-divided from the base; branches

compressed, dilated at the apex into almost a membrane; branchlets minute, irregularly extended and then fimbriate. Color alutaceous red. Does not exceed an inch in height.

The specimen in Schweinitz Herb. is compressed, not fleshy when moistened, and has the hymenium fuscous; basidia longitudinally septate; spores hyaline, even, flattened on one side,  $9 \times 5\frac{1}{2} \mu$ . T. tenax has somewhat the aspect of some forms of T. pallidum but is readily separable from the latter by the very dark hymenium of T. tenax.

Specimens examined:

Massachusetts: West Roxbury, Miss A. Hibbard, under the name T. Hibbardi (in Mo. Bot. Gard. Herb., 58736).

Pennsylvania: Bethlehem, Schweinitz, type (in Herb. Schweinitz and Farlow Herb.).

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# THE IDENTIFICATION OF POLLEN FROM SO-CALLED "HAY FEVER" PLANTS

#### GEORGE T. MOORE

Director of the Missouri Botanical Garden
Engelmann Professor in the Henry Shaw School of Botany of
Washington University

AND

#### ROLAND V. L. LA GARDE

Scientific Assistant, Missouri Botanical Garden

Pollen grains have been the subject of investigation since the earliest days of microscopical examination of plants. References concerning them may be found as early as the end of the seventeenth century, probably the first reference on this subject being that of Marcello Malpighi (1686) in his famous work 'Anatome Plantarum.' Figure 188, on plate 31, shows drawings of pollen grains of a lily which prove the accuracy of observation of the author. In the accompanying description Malpighi says: "\* \* \* Stamineos loculos, globulorum congerie, quasi atomarum, turgere diximus: Hi diversè configurantur, et colorantur, frequentérque luteum sapiunt colorem, ut in lilio, rosis, et limoniis malis; albescunt verò, diaphaníque ferè sunt in malva, et plantagine. Diversâ pariter donantur figurâ in lilio croceo, montano albo. \* \* \* "

From Malpighi up to the beginning of the nineteenth century many references and descriptions of pollen grains are found. Grew pointed out the polymorphism of the pollen grains. Geoffroy spoke of the constancy of size and shape within the species; Needham observed the changes of pollen grains in water preparation, etc. Very correct drawings of pollen grains appear in Purkinje's (1830) work, where the natural classification of plants is attempted and the pollen grains of each group of plants shown. Von Mohl (1835) gives a very complicated and artificial classification of pollen grains based on different marks of identification. An exhaustive citation of the literature from the time of Malpighi is to be found here. Smith ('76) described and drew the pollen grains of a number of plants, and Anderson-Henry ('76) described the pollen grains of two species of Fuchsia in connection with remarks about their hybridization. Edgeworth ('77), Hansgirg ('97), and others made the morphology and anatomy of pollen grains the subject of special investigation, but these authors discussed chiefly European or greenhouse plants which have no direct bearing on the subject in question.

Later there were published a number of papers dealing with the pollen of plants, in which especial attention was paid to so-called "hay fever plants." Among these may be mentioned Scheppegrell's ('22) book, in which he gives a very exact description of the most important hay-fever plants and their pollen. The work is supplemented by many photographs of both the plants and the pollen grains and is certainly of great assistance in the identification of the same. Of some value in identifying pollen grains of hay-fever plants may be mentioned the work of Pope ('25) Koessler and Durham ('26), Waring ('26), Wodehouse ('26), and a report published by the Arlington Chemical Company ('25). Especially in Miss Pope's paper are pollen shapes described, as well as markings, size, color, stickiness, and other characters of importance.

Because of the demand from physicians for a more accurate and definite method than existed for the identification and occurrence of pollen in the vicinity of St. Louis, there was begun in the graduate laboratory at the Missouri Botanical Garden investigations of the pollen of some fifty-five plants regarded as responsible for hay fever. The investigations have been carried on along morphological and microchemical lines for the purpose of devising a key for the identification of pollen grains which occur in the respiratory organs. The results of this work are given herewith in synoptical tables and in a key which it is hoped may

be of some help to botanists and medical men having occasion to identify pollen grains. Attention should be called to the fact that the reactions given may not be typical for pollen grains taken from the respiratory organs or the mucous membrane. It is certain that changes occur in the chemical composition of pollens during their contact with such parts of the body and therefore the chemical reactions noted may be of little or no value under such circumstances. For this reason two different keys have been elaborated; one based on morphological characters and the occurrence of starch, and the other in the form of synoptical tables both morphological and chemical, for the more accurate identification of fresh material.

#### **Methods**

The pollen was either taken from mature flowers and tested the same day, or branches with the ripe flower buds were placed in the incubator until the buds opened, when the fresh pollen was examined. All mounts were made in water. After determining the size and shape of the pollen grains they were stained with "Acid Nigrosine" to determine the number of pores and the presence or absence of lids, after which the other chemical tests were applied.

In all cases the reagent was added in small quantities to the water mounts, except where the pollen had to be tested directly in the reagent in question (i. e., Millon's reagent). Here strong chemicals, such as concentrated acids or lyes, were added drop by drop to avoid a too rapid reaction. In every case the reactions were watched for a long time, or repeated. Tests made from dried herbarium material after it had been kept for years showed that it is possible to identify pollen grains morphologically, the size, shape, and even the number of pores being readily determined. It was not, however, found possible to apply chemical tests to dried pollen, since practically none of the reactions occurring on fresh material took place on the dried specimens.

All investigations and microchemical tests were made with a Zeiss microscope, objective "D" and "ocular No. 3."

#### SHAPE

The most common shapes of the pollen grains investigated were either spherical or elliptical. Tetrahedral and polyhedral grains also occurred, these outlines being modifications of a sphere caused by the pores producing an angular surface. Mounted in water these shapes are especially distinct, due to the difference in swelling of the pollen wall and the pores.

The pollen of *Pinus austriaca*, like that of most of the conifers, is characterized by the presence of two sac-like projections. These air-sacs are apparently nothing more than the enlarged covers of the pores, the grain itself being typically elliptical (see "Acid Nigrosine" below).

#### SIZE AND COLOR

The prevailing size of the pollen grains examined varied from 15 to 40  $\mu$  in diameter. A few were larger, such as Ailanthus glandulosa, 20–50  $\mu$ , Pinus austriaca (without air-sacs), 40–55  $\mu$ , Taraxacum officinale, 35–50  $\mu$ , Polygonum persicaria, 48–70  $\mu$ . For each species the size of the pollen grains is constant within the limit given.

The prevailing color of the pollen grains investigated was yellow; in a few cases, noted in the tables, a brown, gray-yellow, or greenish yellow color occurred. The color is of no diagnostic significance, however, and no use is made of it in the key.

#### SURFACE MARKINGS

Surface markings are important in distinguishing pollen grains, especially if the pores or the thinner places in the pollen wall are considered. Even ignoring the pores, the pollen surfaces of the different species show marked differences. While many are smooth, others have a spiculated or warty surface with spines of different kinds and shapes, with irregular protuberances, reticulations, oil drops, etc.

When the pores are not distinctly visible, they can be brought out more distinctly by certain acids. Reference to such reactions will be found in the synoptical tables. In general, two kinds of pores may be distinguished, large ones and very fine ones, but occasionally there may be a combination of both sizes, so that the pollen may be classified under one of three heads. The large pores may be covered with a distinct lid or closed only with a very delicate membrane, whereas the fine pores are closed with the delicate membrane only. Consequently, according to the nature of the pore covering, a different effect is obtained by treating with certain chemicals or stains. Many of the well-known stains were tried, but no single one was satisfactory for the different pollens. Therefore there was devised a combination of acetic acid and nigrosine, described later under the heading "Acid-Nigrosine," which gave satisfactory results in all cases.

#### THE EFFECTS OF DIFFERENT REAGENTS ON POLLEN GRAINS

Pollen grains have been tested with a series of chemicals and stains commonly used in botanical microtechnics. Some of the reactions are listed in the synoptical tables. During the course of this investigation it has been ascertained that only the tests with three mineral acids (sulphuric, nitric and hydrochloric acids), three alkalies (ammonia water, sodium hydroxide, and potassium hydroxide), and iodine solutions (iodine water and "Lugol's solution") gave reactions which are of any value for identification. Of staining solutions, safranine, fuchsine, methylene blue, gentian violet, and neutral red have been used. The tests with mineral acids gave in general color reactions. In some cases, mentioned in the tables, the appearance of the whole structure or of parts of it was changed.

In concentrated sulphuric acid the contents and the intine are dissolved; usually the dissolved contents swell and creep out through the pores, or when the swelling takes place very rapidly the pollen grains burst. The color reactions in sulphuric acid are mostly confined to the wall. In cases where the contents showed a reaction different from that of the pollen wall it has been mentioned in the tables.

Artemisia absinthium, Aster novae Angliae, and Helianthus annuus have, either in the contents or in the oil drops occurring on the surface, certain yellow or orange-colored pigments. These pigments gave in concentrated sulphuric acid the typical reaction of lipochromes; the natural yellow color turned into green and finally into dark blue, and the contents and oil drops showed

	<del></del>		,	<del></del>	7							
	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec
Acer Negundo												
Acer platanoides	İ			_	_						1	l
Agrostis alba									,			
Ailanthus glandulosa							-					1
Amaranthus retroflexus								_				İ
Ambrosia artemisiaefolia												Ì
Ambrosia trifida										-		
Anthoxanthum odoratum												İ
Artemisia absinthium									-			1
Aster novae Angliae												
Betula populifolia												
Carya alba												
Chenopodium album												
Cheno podium ambrosioides						_						
Chrysanthemum leucanthemum												
Corylus americana		_										
Cynodon Dactylon												
Dactylis glomerata							_					
Dahlia variabilis											1	
Erigeron canadensis												
Festuca elatior								. 1				
Frazinus americana												
Gleditschia triacanthos					1							
Helianthus annuus												
Iva ciliata		1										
Iva xanthifolia						[						
Liquetrum vulgare		1	1									
Liquidambar styraciftua					]		l	j	- 1			
Lolium perenne	Į		ļ		l		_		1			
Medicago sativa	į	- 1					_			_		
Melilotus alba	1	1	1									
Morus alba	1		- 1	_			1			ı		
Panicum anceps			1		- 1	1				i		
Phleum pratense	1	1	- 1		1	I						
Pinus austriaca		1							- i	- 1	İ	
Plantago lanceolata	1	1	- 1			1					ł	
Plantago major	1	1	1								Ì	
Platanus occidentalis	- 1	1		- 1	1						1	
Poa pratensis			- 1	i			- 1		- 1	- 1		
				1						•	- 1	
Polygonum persicaria	- {		- 1	í			- 1				- 1	
Populus balsamifera			1	I	1			- 1	- }			
Pyrus Malus						1	1					
Quercus alba		- 1		Ī						1	- 1	
Quercus coccinea			1	1		1	1				- 1	
Quercus rubra	- 1	Į		ı			1				- 1	
Robinia pseudacacia		- 1	į	-+	-	}				- 1	1	
Rudbeckia laciniata		- 1		1		- 1	t	-		- 1		
Rumex acetosella		1	- 1	ł						- 1		
Solidago canadensis	- 1		- 1		1	1	1				1	
Taraxacum officinale		+	-									
Trifolium pratense	- 1		ł		-	-		$\neg \neg$	$\neg \uparrow$			
Typha latifolia			- 1		ı	1	l	- 1			1	
Ulmus americana	1				- 1	- 1		1	i	1	1	
Kanthium spinosum			- 1		- 1	l	- 1	-		- 1		
Zea Mays			ł	- 1	ŀ	-1	-		- 1		- 1	

small blue granules or crystals. This reaction is very distinct and specific for the pollen of these three plants, and it can be used as a mark of identification.

In nitric and in hydrochloric acid the reaction was chiefly one of color, although, as might be expected, swelling took place in a majority of cases. In nitric acid in two cases, *Chenopodium album* and *Liquidambar styraciflua*, an effect upon the pores was visible, the latter standing out in bold relief. Only in a few instances was it possible to conclude from the results obtained the presence of specific chemical compounds.

With alkalis very little difference was observed between the reaction of the pollen grains of different plants. In the tables a color reaction is indicated, not only of the contents but the separate parts of the pollen grains (perine, extine, intine, and pores) sometimes showing different colors.

"Lugol's solution" and iodine water are used in botanical microtechnics as reagents for proteins and as a test solution for starch. Where starch occurs it turns more or less rapidly to dark blue or black. Among the fifty-five kinds of pollen investigated, in thirty-four cases the presence of starch grains could be determined unquestionably. In twenty-one cases the result was negative (see tables). The size of the starch grains varies from very small to large, a distinguishing mark of some value. In some pollens only a very few starch grains can be traced; other pollen grains appear packed with starch. Pollen from dried material in the herbarium failed to show any starch reaction.

The other reagents with which the pollen grains have been tested were the following: ethylic alcohol (95 per cent), acetone, acetic acid, chromic acid, Biuret's and Millon's reagent, vanilline, aniline sulphate, and diphenylamine. In acetic and chromic acid no changes took place, except in one case. It is of interest that in chromic acid (1 per cent) in one case (Agrostis alba), a swelling of perine and extine occurred. In the other reagents, except Millon's reagent, no change took place which is worth considering.

In Millon's reagent the pollen grains of Ambrosia artemisiaefolia, Chenopodium album, Cynodon Dactylon, Dahlia variabilis, Quercus

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alba, and Q. coccinea gave the positive reaction for proteins. This result is significant for the reason that the effect of proteins is considered to be important in the etiology of hay fever. Six plants out of fifty-five is only about 11 per cent; and of these six plants only two are commonly considered to be hay-fever plants of any great importance, Ambrosia artemisiaefolia and Chenopodium album.

#### "ACID NIGROSINE" STAIN

While some reagents (acids or alkalis) may bring out the pores for a short time, the reaction is not as definite as it should be, particularly where it is desirable to determine the presence or absence of lids. It has been found necessary for quick identification to make the pores distinct with the aid of a stain. In general the pores do not appear, or they are not easily distinguished in water preparations. Repeated experiments have demonstrated that nigrosine combined with an acetic solution produces the best results, since it immediately makes distinct the acid characters desired and the reaction persists for the necessary length "Acid Nigrosine" is made as follows: To 100 cc. of a 2 per cent acetic acid solution is added 5 cc. of a saturated and filtered alcoholic (ethyl) solution of nigrosine (Gruebler). After mixing thoroughly by shaking, the reagent is ready for use. This reagent has proved to be a very helpful one. By means of this stain there can be distinguished four different classes of openings in the pollen wall.

- (1) Pollen grains with large pores which are covered with a lid.
- (2) Pollen grains with large pores without a lid (with a very delicate membrane).
- (3) Pollen grains with large and very small pores (the large with or without lids).
- (4) Pollen grains with a great number of very small pores.

In case 1, the lids of the pores act as a filter. The acid passes through the lids into the contents while the lids hold back the nigrosine and retain it; the lids therefore appear dark violet.

In case 2, where there is no lid (filter), the very delicate membrane allows the stain to pass through unfiltered and to penetrate the contents of the pollen grain. Instead of the contents stain-

ing evenly, the stain takes a definite outline from the opening to the center of the pollen grain in the form of a cylinder or cone. When the stain has reached the center of the pollen grain as many cylinder- or cone-shaped stained areas can be distinguished as there were openings in the pollen wall. Later the stain may spread throughout the contents of the grain, but not until the cylinders or cones above referred to can be readily distinguished, and it is always possible to identify the number of openings in the pollen wall by counting the number of cylinder- or coneshaped regions.

In case 3, the lids of the large pores show the same reaction as in case 1, the stain being retained in the lids. At the same time, however, the stain enters through the small pores and slowly penetrates the contents. The contents beneath the large pores remain unstained, and we are able to count in this way the number of the large pores. If the large pores are not covered with a lid the stain will enter through both the large and small pores, but more rapidly through the large ones because of the larger surface. Since the stain penetrates the contents through the large pores more quickly than through the small ones this makes it possible to determine easily the number of the large pores. Later the contents are stained uniformly.

In case 4, with numerous fine pores, the stain will enter the contents at many places and make its way rapidly and uniformly throughout the interior so that in a short time the whole contents are evenly stained. In some cases a light coloring of the pollen wall may take place but only after the staining of the contents. It is necessary, to obtain the best results, that only small quantities of the stain be applied. Only a small drop should be added to the water preparation and the reaction watched carefully. By using this method and watching the reaction it is easy to determine the kind and number of openings in the wall of the different pollen grains investigated thus far.

The only exception to be noted is that of the pollen of *Pinus austriaca*. The use of "Acid Nigrosine" reagent did not at first seem to demonstrate either pores or lids. After a time, however, the air-sacs began to take the stain, finally assuming a dark violet color. Comparing this result with those obtained with

other pollens, it would seem that the tissue of the air-sacs takes the nigrosine in the same way that the lids of the pores in other pollens do. The color is stored in the air-sac tissue while the acid penetrates the contents. It is possible that the tissue of the air-sac should be regarded as nothing but an enlarged lid of a pore which is concealed below the air-sac in the pollen wall. Since pine pollen has two air-sacs we may conclude that most probably the pollen grains of *Pinus austriaca* have two pores covered with lids which have expanded to produce the air-sacs.

Through the coöperation of Dr. H. L. Alexander, of the Washington University School of Medicine, and with the assistance of Mr. L. B. Harrison, a start was made during the summer of 1926 towards a survey of the air in and around St. Louis. By this means, if the work can be continued through several seasons, it is hoped that an accurate knowledge of the prevailing pollens in the air at different times may be obtained.

Various stations were established by Mr. Harrison, and ordinary microscope slides, covered with a film of cotton-seed oil, were exposed for twenty-four hours. These were then brought to the laboratory and examined under the microscope. An attempt was made to use the Cohen dust pump for collecting pollen from the air but the apparatus proved to be too perfect in that it gathered in so much dust with the pollen that the pollen could not be identified. On the whole, no more satisfactory method of getting samples of the pollen in the air than by the exposure of plates covered with some oil or similar adhesive material has been developed.

The observations extended over a period of 115 days (from June 23, 1926, to October 15, 1926). Owing to relative absence of pollen on some days, rain or other disturbing factors, only on 51 days were there positive results. These, with the station from which obtained and the name of the plant, are given below.

Date	Station*	Plant
June 23	1	Chenopodium ambrosioides
June 26	2	Phleum pratense
June 27	1 3	Agrostis alba, Dactylis glomerata Dactylis glomerata, Plantago lanceolata
June 28	2 3	Dactylis glomerata, Agrostis alba Dactylis glomerata, Phleum pratense
June 29	2	Phleum pratense, Agrostis alba
June 30	1	Poa pratensis, Phleum pratense
July 1	5	Poa pratensis
July 2	5 1 2	Poa pratensis, Agrostis alba Poa pratensis, Festuca elatior Poa pratensis
July 3	2 3	Phleum pratense, Poa pratensis Plantago lanceolata, Phleum pratense
July 4	2 3	Poa pratensis, Agrostis alba, Phleum pratense Phleum pratense
July 5	1	Festuca elatior
July 6	1 2	Lolium perenne, Rumex acetosella Poa pratensis
July 7	1 3 6	Phleum pratense Phleum pratense Rumex acetosella, Agrostis alba
July 8	2 3 6	Plantago lanceolata, Rumex acetosella Ambrosia artemisiaefolia, Panicum anceps, Festuca elatior Phleum pratense, Poa pratensis
July 11	1	Phleum pratense, Festuca elatior
July 12	2	Chenopodium ambrosioides
July 14	6	Festuca elatior, Chenopodium ambrosioides
July 15	3, 4	Chenopodium ambrosioides, Festuca elatior
July 16	2, 4	Festuca elatior Chenopodium ambrosioides
July 19	3 4	Festuca elatior Plantago lanceolata, Phleum pratense
July 20	1, 2 3 6	Chenopodium ambrosioides Festuca elatior Chenopodium ambrosioides, Festuca elatior
July 23	1 2	Zea Mays Festuca elatior

### ANNALS OF THE MISSOURI BOTANICAL GARDEN

Date	Station*	Plant
July 24	<b>4</b> 6	Phleum pratense Phleum pratense
July 29	1 4	Chenopodium ambrosioides Ambrosia artemisiaefolia
July 31	1, 2	Ambrosia artemisiaefolia
Aug. 1	1	Ambrosia artemisiaefolia
Aug. 2	2	Ambrosia artemisiaefolia
Aug. 3	6	Ambrosia artemisiaefolia
Aug. 4	2 4,6	Ambrosia artemisiaefolia Ambrosia trifida
Aug. 5	6	Ambrosia artemisiaefolia
Aug. 6	1	Zea Mays (?)
Aug. 10	4 6	Phleum pratense Chenopodium ambrosioides, Iva ciliata, Iva xanthifolia
Aug. 11	1 4	Chenopodium ambrosioides, Amaranthus retroflexus Ambrosia artemisiaefolia
Aug. 13	3	Amaranthus retroflexus
Aug. 14	6	Ambrosia artemisiaefolia
Aug. 15	2, 3, 4	Ambrosia artemisiaefolia, Amaranthus retroflexus Ambrosia artemisiaefolia
Aug. 22	2	Ambrosia artemisiaefolia
Aug. 24	6	Chenopodium ambrosioides, Taraxacum officinale
Aug. 25	1-4,6	Ambrosia artemisiaefolia (great quantities)
Aug. 26	$\begin{bmatrix} 1 \\ 2, 6 \\ 3 \\ 4 \end{bmatrix}$	Erigeron canadensis Ambrosia artemisiaefolia Ambrosia artemisiaefolia, Ambrosia trifida Ambrosia artemisiaefolia, Solidago canadensis
Aug. 28	2,3	Ambrosia artemisiaefolia
Aug. 30	6	Ambrosia artemisiaefolia
Sept. 1	1, 3, 4, 6	Ambrosia artemisiaefolia
Sept. 7	2, 4 3	Ambrosia artemisiaefolia Ambrosia artemisiaefolia, Chenopodium album
Sept. 17	1, 2, 3, 4	Ambrosia artemisiaefolia
Sept. 18	3, 4, 6	Ambrosia artemisiaefolia, Chenopodium album Ambrosia artemisiaefolia

Date	Station*	Plant
Sept. 20	3 2 6	Chenopodium album Chenopodium album, Ambrosia artemisiaefolia Ambrosia artemisiaefolia, Solidago canadensis
Sept. 21	4	Ambrosia artemisiaefolia, Chenopodium album, Medicago sativa
Sept. 30	6	Ambrosia artemisiaefolia (great quantities)
Oct. 6	3, 4	Ambrosia artemisiaefolia
Oct. 7-15		No pollen grains

<sup>\*</sup> Index of Stations: 1, # 4700 McPherson; 2 and 3, Skinker Road in front of Fine Arts Building of Washington University; 4, Skinker Road at University Street Car Line; 5, south of Forest Park, east of Forest Park Highlands; 6, Medical School of Washington University.

According to the frequency of pollen grains occurring on the slides, the more important plants can be arranged as follows:

Pollen of	Number of days occurred	Per cent
Ambrosia artemisiaefolia	24	49
Phleum pratense	12	24
Chenopodium ambrosioides	10	20
Festuca elatior	9	18
Poa pratensis	7	14
Agrostis alba	6	12
Chenopodium album, Plantago lanceolata	4	8
Amaranthus retroflexus	4 3	6
Ambrosia trifida, Dactylis glomerata, Rumex acetosella, Soli- dago canadensis, Zea Mays	) 2	4
Erigeron canadensis, Iva ciliata, Iva xanthifolia, Lolium perenne, Medicago sativa, Panicum anceps, Taraxacum officinale	1	2

#### DESCRIPTION OF POLLENS

Acer Negundo (Box Elder). No. 50.1—Pollinating from middle of March until middle of April. Tetrahedral, 3 pores without lids; 25–40 μ. Contents granular. The dry pollen taken directly from the anthers is folded and shaped like a grain of rye; in water preparation the pollen grains stretch and become tetrahedral. Color grayish yellow. In "Acid Nigrosine" the contents below the pores stain dark violet. Starch present.

Acer platanoides (Norway Maple). No. 43.—Pollinating from middle of April until middle of May. Tetrahedral, 3 pores with

<sup>&</sup>lt;sup>1</sup> Numbers refer to those used in key.

lids;  $30 \times 38$   $\mu$ . Contents coarse-grained. The pollen wall very thick and shows distinct layers. Color yellowish gray. "Acid Nigrosine" stains the lids of the pores dark violet. No starch present.

Agrostis alba (White Bent Grass). No. 28.—Pollinating May and June. Elliptical, 1 pore with a lid;  $17 \times 24-25 \times 32~\mu$ . Contents finely granulated. Pollen wall thick. Extine with very delicate light red-brown shimmer. In water grains appear bean-shaped. Colorless. "Acid Nigrosine" stains first the lids of the pores and later the contents. In chromic acid the wall swells and appears very thick (3.5  $\mu$ ). Starch present.

Ailanthus glandulosa (Tree of Heaven). No. 45.—Pollinating middle of May until end of June. Tetrahedral, in water more or less spherical, 3 large pores with lids and numerous fine pores;  $20{\text -}50~\mu$ . Contents finely granular. Color yellow. "Acid Nigrosine" stains the lids of the pores dark violet. Starch may or may not be present. In ammonia water the fine pores become very distinct.

Amaranthus retroflexus (Pigweed). No. 11.—Pollinating end of July until middle of August. Spherical, numerous fine pores with lids in the thick wall; 23–25 µ. Color grayish yellow. "Acid Nigrosine" stains the pores dark blue and later the contents become violet. Starch present.

Ambrosia artemisiaefolia (Common or Lesser Ragweed). No. 18.—Pollinating beginning of August until middle of October. Spherical, 3 pores with lids. Pores placed equatorially. Surface studded with obtuse spines; 17–22  $\mu$ . Dry grains appear compressed and therefore elliptical, but in water they stretch and become spherical. Color yellow. "Acid Nigrosine" stains only the lids of the pores dark violet. No starch present. Contents give very weak protein reaction in both Millon's and Biuret's reagent.

Ambrosia trifida (Great Ragweed). No. 27.—Pollinating August and September. Spherical, 3 pores without lids. Pores placed equatorially. Surface studded with short obtuse spines; 35–42  $\mu$ . Color golden. "Acid Nigrosine" stains the pores dark violet. Starch present.

Anthoxanthum odoratum (Sweet Vernal Grass). No. 29.—Pol-

linating first half of May. Elliptical, almost spherical, 1 pore with lid;  $32\times25$ – $34\times37$   $\mu$ . Contents coarse-grained. Color yellowish. "Acid Nigrosine" stains the lid of the pore dark violet. Starch present.

Artemisia absinthium (Wormwood). No. 12.—Pollinating June until the end of August. Spherical, 3 pores with lids. Spiculated surface with oil drops; 20–28 µ. The dry grains appear elliptical. Color yellow. "Acid Nigrosine" stains the lids of the pores dark violet. No starch present. Sulphuric acid produces in the dissolved contents blue granules and crystals (lipochromes?).

Aster novae Angliae (New England Aster). No. 13.—Pollinating August until October. Spherical, in water somewhat flat, 3 pores with lids; 24–28  $\mu$ ; young grains 17–22  $\mu$ . The surface is studded with short warty spines, 1.7–3.5  $\mu$  in length, covered with numerous oil drops. Color yellow. In "Acid Nigrosine" the lids of the pores swell and stain dark violet. In mature grains the extine separates from the pores and they stand out in bold relief. No starch present. In sulphuric acid the dissolved contents and the oil drops appear blue (lipochromes).

Betula populifolia (American White Birch). No. 7.—Pollinating end of February to the middle of March. Spherical, 3 pores with lids;  $27-32~\mu$ . Color yellow. "Acid Nigrosine" stains the lids of the pores dark violet. Abundant starch present.

Carya alba (Shagbark Hickory). No. 34.—Pollinating last half of May. Elliptical, 3 pores (sometimes 4) with lids;  $38 \times 49$ – $63 \times 70~\mu$ . Pollen wall thick (3.5  $\mu$ ). Light grayish yellow. "Acid Nigrosine" stains lids of the pores dark violet. In acetic acid the pollen grains become spherical and show fine granulated contents with large oil drops. Starch present.

Chenopodium album (Lamb's Quarters). No. 26.—Pollinating July until the end of September. Spherical, numerous small pores without lids; 20–32 µ. Surface smooth, contents coarsegranular. Color dirty yellow. "Acid Nigrosine" stains first the pores dark violet, later the contents. Starch present. Millon's reagent gives a positive result.

Chenopodium ambrosioides (Wormseed). No. 55.—Pollinating end of August until end of October. Polyhedral, numerous fine pores; 24–28 µ. Surface appears uneven, contents coarse-

granular. Color yellowish gray. "Acid Nigrosine" rapidly stains the contents violet. In acetic acid the pores become distinctly visible. Starch present.

Chrysanthemum leucanthemum (Ox-eye Daisy). No. 47.—Pollinating July until the middle of August. Tetrahedral, 3 pores with lids; 23–28 µ. Surface studded with spines, contents granulated. "Acid Nigrosine" stains lids of pores dark violet. Color yellowish gray. No starch present.

Corylus americana (American Hazelnut). No. 44.—Pollinating end of February and first half of March. Tetrahedral, 3 pores with lids; 28–35 µ. Surface smooth, contents granulated. Color yellow. "Acid Nigrosine" stains lids of pores dark violet. Methylene blue stains first the pores dark blue, then the contents. Starch present.

Cynodon Dactylon (Bermuda Grass). No. 1.—Pollinating June until middle of September. Spherical, 1 pore with a lid;  $30-38~\mu$ . Contents granulated, surface smooth. "Acid Nigrosine" stains the lid of the pore dark violet. Color dirty yellow. Starch present. Millon's reagent gives a positive result.

Dactylis glomerata (Orchard Grass). No. 30.—Pollinating end of May and during June. Elliptical, 1 pore with a lid;  $24 \times 28$ –  $32 \times 39$   $\mu$ . Surface smooth, contents coarse-granular. Color grayish. "Acid Nigrosine" stains first the lids of the pores and then the perine. Abundant starch present.

Dahlia variabilis (Common Dahlia). No. 21.—Pollinating September until October (first frost). Spherical, 12 to 20 large pores with lids; 28–35  $\mu$ . Surface covered with oil drops and studded with sharp-pointed spines 3.5  $\mu$  in length. Color yellow. "Acid Nigrosine" stains the lids of the pores dark violet. Starch present. Sodium hydroxide changes the color of the contents from bright red to orange, then light brown and finally to yellow (typical tyrosine reaction). The perine swells. Millon's reagent gives a positive result.

Erigeron canadensis (Horseweed). No. 15.—Pollinating August and September. Spherical, 3 pores with lids; 16–22 μ. Surface spiny. Color pale yellowish gray. "Acid Nigrosine" slowly stains the lids of the pores violet, gradually becoming darker; later the entire contents become dark violet. No starch present.

Festuca elatior (Meadow Fescue). No. 2.—Pollinating June and July. Spherical, 1 pore with a lid; 23–30 μ. Colorless. "Acid Nigrosine" stains the lid of the pore dark violet, later the contents light violet. Starch present.

Fraxinus americana (White Ash). No. 38.—Pollinating second and third week in April. Elliptical, 3, sometimes 4, pores without lids, the pores arranged in a circle around the longer axis;  $21 \times 24-24 \times 32 \mu$ . Surface smooth, contents granular. Color brown. "Acid Nigrosine" stains the contents beneath the pores dark violet. Starch present.

Gleditschia triacanthos (Honey Locust). No. 35.—Pollinating from the middle until the end of May. Elliptical, almost spherical, 3 pores with lids;  $28 \times 32-42 \times 46 \mu$ . Surface finely granular with oil drops in places. Color light greenish yellow. In "Acid Nigrosine" the lids of the pores swell and stain dark violet. No starch present.

Helianthus annuus (Common Sunflower). No. 14.—Pollinating July until end of September. Spherical, 3 pores with lids; 30–40  $\mu$ . Surface covered with oil drops and studded with sharp-pointed spines, 3.5–7  $\mu$  in length. Color dirty yellow. In "Acid Nigrosine" lids of pores swell and stain dark violet. In sulphuric acid the oil drops turn blue (lipochromes?). No starch present.

Iva ciliata (Rough Marsh Elder). No. 17.—Pollinating August until middle of October. Spherical, 3 pores with lids; 24–28  $\mu$ . Surface spiny, wall very thick. Color grayish yellow. "Acid Nigrosine" stains dark violet, first, the lids of the pores, later, the contents. Very few starch grains.

Iva xanthifolia (Burweed Marsh Elder). No. 41.—Pollinating July until September. Elliptical, 3 pores without lids;  $14\times17$ – $17\times21~\mu$ . Surface studded with spines. Color grayish yellow. With "Acid Nigrosine" the stain enters the pores and penetrates the contents to the center, forming dark violet-colored cones. Starch present.

Ligustrum vulgare (Privet). No. 54.—Pollinating May until July. Spherical, 3 (sometimes 4) pores without lids; 24–35  $\mu$ . Surface reticulated. Color yellow. Contents granulated. "Acid Nigrosine" stains the contents below the pores dark violet. No starch could be recognized.

Liquidambar styraciflua (Sweet Gum). No. 10.—Pollinating second half of April. Spherical, 12–20 large pores with lids and numerous fine pores; 35–42  $\mu$ . Surface smooth, wall very thick (1.7–3.5  $\mu$ ). Color yellowish. "Acid Nigrosine" stains the lids of the large pores, and since the stain enters through the small pores, the contents in a short time appear dark violet. If only small quantities of the stain are used the larger pores can easily be detected. In chloral hydrate the structure of the grains becomes distinct, especially the pores and the layers of the wall, and the grains assume a polyhedral shape. Starch present.

Lolium perenne (Darnel or Rye Grass). No. 31.—Pollinating last week of May until end of June. Elliptical, 1 pore with a lid;  $24 \times 31-32 \times 39 \mu$ . Surface smooth, wall very thick (2.5-3  $\mu$ ). Color yellowish. Contents granular. Lid of the pore stains dark violet in "Acid Nigrosine." No starch present.

Medicago sativa (Alfalfa). No. 36.—Pollinating May until October. Elliptical, 3 pores with lids;  $31 \times 35-42 \times 45 \mu$ . Surface reticulated. Color grayish yellow. Contents granular. "Acid Nigrosine" quickly stains the lids of the pores dark violet. No starch present.

Melilotus alba (Sweet clover). No. 48.—Pollinating May until October. Tetrahedral, 3 pores with lids; 20–28 μ. Surface with cone-shaped projections from the pores. Color greenish yellow. Contents granular. "Acid Nigrosine" stains the lids of the pores at first, later the projections. No starch present.

Morus alba (White Mulberry). No. 6.—Pollinating second half of April. Spherical, 2 pores with lids; 17–21 μ. Surface smooth. Contents granular. Color light gray-brown. "Acid Nigrosine" acts slowly, staining first the lids of the pores and later the pollen wall. Starch present.

Panicum anceps (Beaked Panicum). No. 3.—Pollinating early July until September. Spherical, 1 pore with lid (dry pollen grains appear elliptical). The pore stands out in bold relief; 30–38  $\mu$ ; Surface smooth. Contents granular. Color light yellowish gray. "Acid Nigrosine" stains the lid dark violet. Starch present.

Phleum pratense (Timothy). No. 32.—Pollinating June until end of August. Elliptical, 1 pore with lid;  $25 \times 31-33 \times 35 \mu$ .

Surface smooth. Contents granular. Color light yellow. "Acid Nigrosine" quickly stains the lids of the pores; later the contents are faintly colored. Starch present.

Pinus austriaca (Austrian Pine). No. 40.—Pollinating first half of May. Elliptical (swelling in water until almost spherical), with 2 air-sacs and probably with 2 pores;  $31 \times 50-50 \times 70~\mu$ ; without air-sacs,  $31 \times 40-38 \times 55~\mu$ . No surface markings except air-sacs. Color light yellow with air-sacs black. "Acid Nigrosine" slowly stains the air-sacs violet. In sulphuric acid the air-sacs show a very fine reticular structure. In acetic acid the grains swell and the wall appears very thick  $(5.2~\mu)$ ; the pollen grains become bean-shaped. Contents granular. Starch present.

Plantago lanceolata (English Plantain or Rib Grass). No. 9.—Pollinating middle of May until end of September. Spherical, 12 pores with lids; 20–28 μ. Surface smooth. Contents coarsely granular. Color yellowish gray. "Acid Nigrosine" stains lids of pores dark violet. Starch present.

Plantago major (Common Plantain). No. 8.—Pollinating June until end of September. Spherical, 6 pores with lids; 20–25 μ. Surface smooth, wall thick. Contents coarsely granular. Color very pale yellow, almost colorless. "Acid Nigrosine" stains at first the lids of the pores and later the contents. Starch present.

Platanus occidentalis (Sycamore or Buttonwood). No. 51.—Pollinating in May. Tetrahedral, 3 pores without lids; 20–34  $\mu$ . Surface smooth. Contents coarsely granular. Color weak dirty yellow. "Acid Nigrosine" stains at first beneath the pores, later all the contents. Starch grains very small and not in great quantity.

Poa pratensis (Blue Grass). No. 4.—Pollinating from middle of May until end of September. Spherical, 1 pore with a lid;  $31-37~\mu$ . Surface smooth. Contents coarsely granular. Color greenish yellow. In "Acid Nigrosine" the lids of the pores first stain dark violet, whereas the pores themselves appear greenish blue. Later the contents take the color and stain dark violet. No starch present.

Polygonum persicaria (Knotweed or Lady's Thumb). No. 23.—

Pollinating August and September. Spherical, numerous large pores (more than 20) with lids; 28–53  $\mu$ . Surface reticulated. Contents granular. Color very light yellow. "Acid Nigrosine" stains the lids of the pores dark violet. In sulphuric, nitric, and hydrochloric acid the folded structure of the reticulations becomes very distinct. Starch present.

Populus balsamifera (Balsam Poplar). No. 25.—Pollinating middle of March. Spherical, numerous small pores; 20–40 μ. Surface fine-granular. Contents coarse-granular. Color yellow. "Acid Nigrosine" stains the contents dark violet, the wall light violet. The intine and likewise the contents give a positive reaction for myriophylline; in vanilline-hydrochloric acid, pinkish and purple; in diphenylamine from yellow to pink to brown. Raciborski ('93) has determined myriophylline in the young leaves of Myriophyllum (hence the origin of the name). Starch present.

Pyrus Malus (Common Apple). No. 52.—Pollinating in May. Tetrahedral, 3 pores without lids; 34–35 μ. Surface marked with cone-shaped projections from the pores. Contents coarse-granular. Color pale yellowish gray. "Acid Nigrosine" stains the contents dark violet immediately beneath the pores; later the stain moves toward the center in cone-shaped areas. No starch present.

Quercus alba (White Oak). No. 19.—Pollinating in May. Spherical, 3 pores with lids which are elongated in cone-shaped projections; 28–34 μ. Contents coarse-granular, containing, in addition to a few small starch grains, globoids with protein crystals. Wall thick. Color yellowish. "Acid Nigrosine" stains the lids of the pores and the projections from the lids dark violet. In potassium hydroxide the globoids become distinct. The contents give a positive protein reaction in Millon's reagent. Starch present.

Quercus coccinea (Scarlet Oak.) No. 49.—Pollinating in May. Tetrahedral, 3 pores with lids and cone-shaped projections from the pores; 24–35  $\mu$ . Surface smooth, wall thick. Contents coarse-granular. In "Acid Nigrosine," at first the lids of the pores stain dark violet, then the projections light violet; later the places beneath the pores take the color. The contents give

the positive protein reaction in Millon's reagent. No starch present.

Quercus rubra (Red Oak). No. 42.—Pollinating in May. Tetrahedral, 3 pores with lids; 24–35  $\mu$ . Surface smooth. Contents coarse-granular. Color dirty yellow. "Acid Nigrosine" stains the lids of the pores dark violet, later penetrating beneath the pores. No starch present.

Robinia pseudacacia (Common Locust). No. 46.—Pollinating at end of April and early in May. Tetrahedral, 3 pores with lids; 28-41 µ. Surface finely granulated. Color light grayish yellow. "Acid Nigrosine" stains the lids of the pores dark violet. Starch present in very small grains.

Rudbeckia laciniata (Tall Cone-flower). No. 16.—Pollinating in August and September. Spherical, 3 pores with lids; 20–25  $\mu$ . Surface studded with spines about 3.5  $\mu$  in length. Color yellow. "Acid Nigrosine" stains the lids of the pores dark violet. No starch present.

Rumex acetosella (Sheep Sorrel). No. 39.—Pollinating from May until August. Elliptical, 4 pores without lids;  $21 \times 24-24 \times 28$   $\mu$ . Surface smooth with occasional oil drops. Contents coarsely granular. Color yellow. "Acid Nigrosine" very slowly stains the pores pale violet.

Solidago canadensis (Canada Golden-rod). No. 37.—Pollinating from August until first half of October. Elliptical, 3 pores with lids;  $15 \times 21-18 \times 24$   $\mu$ . Surface studded with obtuse spines and covered with oil drops. The spines are arranged in rows parallel to the longer axis. "Acid Nigrosine" stains the lids of the pores dark violet. No starch present.

Taraxacum officinale (Dandelion). No. 22.—Pollinating all the year. Spherical, 12–20 pores with lids; 35–50 μ. Surface reticulated and studded with short blunt spines. Oil drops also abundantly present. Color gold. "Acid Nigrosine" very slowly stains the pores. Sulphuric acid turns the oil drops blue (lipochromes?). No starch present.

Trifolium pratense (Red Clover). No. 20.—Pollinating from April until November. Spherical, 3 pores with lids and coneshaped projections from the pores; 28-41 µ. Surface smooth. Color grayish. "Acid Nigrosine" stains the pores and the lids dark violet, the projections light violet. No starch present.

Typha latifolia (Common Cat-tail). No. 5.—Pollinating in June. Single pollen grains spherical, 1 pore with a lid with margin, pollen wall penetrated by very fine pores; 24–35  $\mu$ ; groups from 2–5 pollen grains  $35\times42$ –49  $\mu$ , and  $38\times42$ –45  $\mu$ . Single pollen grains occur rarely; usually they are united in irregular aggregations from 2–5 grains. Surface smooth. Color yellow. "Acid Nigrosine" stains only the lids of the pores dark violet. In sulphuric acid the structure of the extine becomes very distinct and the numerous fine pores can be identified very easily. In contradistinction to Typha latifolia, Typha angustifolia has only single pollen grains which never occur in aggregations. Size 30–42  $\mu$ . Each pollen grain has 1 pore with lid, and numerous fine pores, but the surface is granulated. Starch present.

Ulmus americana (American Elm). No. 53.—Pollinating middle of March until middle of April. Polyhedral, 5 pores with lids; 25–35 μ. Surface smooth. Contents finely granular. Color grayish yellow. "Acid Nigrosine" stains only the lids of the pores dark violet. No starch present.

Xanthium spinosum (Burweed or Cocklebur). No. 24.— Pollinating from second week of August until second half of September. Spherical, 3 large pores without lids and numerous fine pores; 22–28 μ. Surface smooth. Contents coarse-granular. Color light brown. In "Acid Nigrosine" the pores and places beneath the pores stain dark violet; the contents stain later. No starch present.

Zea Mays (Indian Corn). No. 33.—Pollinating latter part of June and first half of July. Elliptical, 1 assymetrically placed pore with a lid;  $70 \times 75\text{--}85 \times 88~\mu$ . The differences in length of the axis are not very great but spherical pollen grains occur rarely. Surface smooth. Contents granular. Color light yellow. "Acid Nigrosine" stains first the lids of the pores dark violet, later the pollen wall. Starch present.

#### KEY

The key is based primarily on the shape of the pollen and number of pores. As subdivisions the presence or absence of lids on the pores and surface markings have been used because they are characteristics and stable. The presence of different projections, such as spines, warts, etc., is very helpful in identification. For some of the spherical pollen grains it was necessary to take into account the presence or absence of starch as well as the comparative size of the grains. Starch, which can be determined easily, is perhaps the only chemical distinction which is of use. Attention should also be paid to the time of pollination which is of great assistance in some cases.

It may be a disputed point whether the tetrahedral and polyhedral pollen should not be regarded as spherical. While these are actually spherical, departure from this shape is caused by the pores, which by their position produce a three- or many-sided appearance. However, since under practically all conditions the angular shape remains constant and there is no difficulty in recognizing it under the microscope, it seems one of the most readily determined characters for identification.

#### KEY

KEY		
I. Spherical pollen grains, large pores with lids		
1. Without surface markings		
A. 1 pore		
a. Single pollen grains		
$\alpha$ . With starch grains 0.5-1.7 $\mu$		
Starch grains 0.5–0.6 $\mu$ ; pollen grains 30–38 $\mu$ .	June-Sept.	Cynodon Dactylon
Starch grains 0.8–1.1 $\mu$ ; pollen grains 23–30 $\mu$	June, July	Festuca elatior
Starch grains 1.1-1.7 $\mu$ ; pollen grains 30-38 $\mu$	July-Sept.	Panicum anceps
β. Without starch grains		
Coarse-grained; pollen grains $31-37 \mu$	May-Sept.	Poa pratensis
b. Pollen grains aggregated in groups from 2-5,		
$35 \times 42-35 \times 49 \mu$ ; single pollen grains 24-	T	Manual - 1-425-12-4
35 µ	June	Typha latifolia*
B. 2 pores 17–21 μ	April	Morus alba
C. 3 pores	Арти	moras aroa
27-32 µ	Feb., Mar.	Betula populifolia
D. 6 pores	2 001, 212021	zama popunyona
20-25 μ	June-Sept.	Plantago major
E. 12 porcs		
20-28 μ	May-Oct.	Plantago lanceolata
F. 12-20 porcs		
$35$ – $42~\mu$	April	Liquidambar styraciflua
G. More than 20 pores	* 1 4	4
$\frac{23-25}{4}$	July., Aug.	Amaranthus retroflexus
2. With surface markings		
A. 3 pores		
a. Surface spiny		
α. Oil drops on surface Spines 1.7–3.5 μ; pollen grains 20–28 μ	June-Aug.	Artemisia absinthium
Spines 1.7-3.5 $\mu$ ; pollen grains 24-28 $\mu$	AugOct.	Aster novae Angliae
Spines 1.7-3.5 $\mu$ , policin grains 24-25 $\mu$ Spines 3.5-7 $\mu$ ; policin grains 30-40 $\mu$	July-Sept.	Helianthus annuus
β. No oil drops on surface	out, tept.	11 CI DOTHING CHINES
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Aug., Sept.

Erigeron canadensis

Aug., Sept. Rudbeckia laciniata

Spines  $0.8-0.9~\mu$ ; pollen grains  $16-22~\mu$ Spines  $3.5~\mu$ ; pollen grains  $20-25~\mu$ 

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b. Surface warty	. 0.1	Same - 27.5 m.k.	
<ul> <li>Warts 1.2 μ; pollen grains 24–28 μ</li> <li>Warts 1.7 μ; pollen grains 17–22 μ</li> <li>c. Surface with cone-shaped projections from pores</li> </ul>	AugOct. AugOct.	Iva viliata Ambrosia artemisiasfolia	17 18
Starch present; pollen grains 28–34 μ Starch absent; pollen grains 28–41 μ	May AprNov.	Quercus alba Trifolium pratense	18 20
B. 12-20 pores a. Surface with oil drops and sharp spines; pollen			
grains $28-35 \mu$ b. Surface with oil drops and short blunt spines;	Sept., Oct.	Da <b>kli</b> a variabilis	21
pollen grains 35–50 $\mu$	All the year	Taraxacum officinale	22
C. Numerous pores a. Surface with hexagonal reticulations; pollen grains 28-53 \( \mu \)	Aug., Sept.	Poly <b>gon</b> um persicaria	23
II. Spherical pollen grains, large pores without lids			
1. Without a surface marking; with numerous fine			
pores A. 3 large pores; contents coarse; pollen grains 22-28 \( \mu \)	Aug., Sept.	Xanthium spinosum	24
B. No large pores Pollinating in March; pollen grains 20-40  Pollinating July-September; pollen grains 20-32   Note of the pollen grains 20-32   District of the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-32   But the pollen grains 20-40   But	March July-Sept.	Populus <b>balsamifera</b> Chenopo <b>dium album</b>	25 26
2. With surface markings A. 3 pores; surface spiculated; pollen grains 35–42 $\mu$	Aug., Sept.	Ambrosia trifida	27
III. Elliptical pollen grains, large pores with lids			
1. Without surface markings A. 1 pore			
<ul> <li>a. Pore placed laterally; very small starch grains; pollen grains 17 × 24-25 × 32 μ</li> <li>b. Pore placed at one end of the longer axis;</li> </ul>	May, June	Agrostis alba	28
pollen grains 32 × 35-34 × 37 μ; small starch grains c. Pore placed at one end of the longer axis; num-	May	Anthoxanthum odoratum	29
erous fine pores in the pollen wall; large starch	May Juna	Dagtulia alumanta	30
grains; pollen grains $24 \times 28-32 \times 39 \mu$ d. Pore placed at one end of the longer axis; no	May, June	Dactylis glome <b>rata</b>	
starch grains; pollen grains $24 \times 31-32 \times 39 \mu$ e. Pore placed laterally; large starch grains; pollen	May, June	Lolium perenne	31
grains $25 \times 31-33 \times 35 \mu$	June-Aug.	Phleum pratense	32
f. Pore placed laterally; large starch grains; pollen grains $70 \times 75-85 \times 88 \ \mu$ B. 3 pores	June, July	Zea Mays	<b>3</b> 3
Starch grains; pollen grains 38 × 49-63 × 70 $\mu$ No starch grains; pollen grains 28 × 32-42 × 46 $\mu$ 2. With surface markings	May May	Carya alba Gleditschia triacant <b>hos</b>	34 35
A. 3 pores			
Small reticulations; pollen grains 31 $\times$ 35–42 $\times$ 45 $\mu$	May-Oct.	Medicago sativa	36
Obtuse spines; pollen grains 15 $\times$ 21–18 $\times$ 24 $\mu$	AugOct.	Solidago canadensis	37
IV. Elliptical pollen grains, large pores without lids			
1. Without surface markings A. 3-4 pores; pollen grains $21 \times 24-24 \times 31 \mu$	April	Fraxinus americana	38
B. 4 pores; pollen grains $21 \times 24-24 \times 28 \mu$	May-Aug.	Rumex acetosella	39
2. With surface markings A. 2 pores; 2 air-sacs; pollen grains $31 \times 50-50 \times 70 \mu$ $(31 \times 40-38 \times 55 \mu)$	May	Pinus austriaca	40
B. 3 pores; surface spiny; pollen grains 14 $\times$ 17-17 $\times$ 21 $\mu$	July-Sept.	Iva xanthifolia	41
/ F	car sopu		**

#### V. Tetrahedral pollen grains

A. S pores with lids 1. Surface smooth			
a. Pollinating in May; contents coarse-grained;			
pollen grains 24–35 $\mu$	May	Quercus rubra	42
<ul> <li>b. Pollinating in April; contents coarse-grained; pollen grains 30-38 μ</li> <li>c. Pollinating in February and March; contents</li> </ul>	April, May	Acer platanoides	43
fine-granulated; pollen grains 28-35 $\mu$ d. Pollinating in May and June; contents fine-	Feb., Mar.	Corylus americana	44
granulated; pollen grains 20-50 $\mu$ 2. Surface with markings	May, June	Ailanthus glandulosa	45
a. Surface fine-granulated: pollen grains 28-41 u	April, May	Robinia pseudacacia	46
b. Surface spiny; pollen grains 23-28 μ	July, Aug.	Chrysanthemum leucan- themum	47
c. Surface with cone-shaped projections from			
α. Contents fine-granular, starch present; pollen grains 20-28 μ	May-Oct.	Melilotus alba	48
β. Content coarse-grained; no starch present; pollen grains 24-35 μ	M	0	40
B. S pores without lids	May	Quercus coccinea	49
a. Surface smooth			
<ul> <li>c. Contents granular; pollen grains 25-40 μ</li> <li>β. Contents coarse; pollen grains 20-34 μ</li> </ul>	Mar., Apr. May	Acer Negundo Platanus occide <b>ntalis</b>	50 51
b. Surface with cone-shaped projections from pores; contents coarse; pollen grains 34-35 $\mu$	May	Pyrus Malus	52
VI. Polyhedral pollen grains			
1. With lids			
A. 5 pores; pollen grains 25-35 μ 2. Without lids	Mar., Apr.	Ulmus americana	53
<ul> <li>A. 3 large pores; pollen grains 24-35 μ</li> <li>B. Numerous fine pores; pollen grains 24-28 μ</li> </ul>	May-July AugOct.	Ligustrum vulgare Chenopodium ambrosi-	54
. /1		oides	55

#### SYNOPTICAL TABLE

As a complement to the key, the following synoptical table has been prepared. The table may be of some assistance to those wishing to have a summary of the various characters of a particular pollen. Only the more important reactions have been noted, it being superfluous to mention reactions which occur universally and are very well known to every investigator.

<sup>•</sup> The pollen of  $Typha\ latifolia$  is easily distinguished from that of  $Typha\ angustifolia$ , the latter having grains always spherical and occurring singly, never in aggregations. Measurements: 30–42  $\mu$ . The surface is fine-granulated and the pollen wall is penetrated by numerous fine pores. Each pollen grain has one large pore with a lid.

Plant	Acer Negundo	Acer platanoides	Agrostis alba	Ailanthus glandulosa
Pollinating	Mar. 23-Apr. 4	AprMay	May-June	End May-end June
Size	25-40 μ	30-38 μ	$17  imes 24$ – $25  imes 32  ext{ }\mu$	20-50 μ
Shape and marks*	t O1; S smooth; C granular	t O <sup>2</sup> L; S smooth; C coarse	e O¹ L; S smooth; C fine-granulated	t O <sup>3</sup> L o <sup>n</sup> ; S smooth; C granular
Color	Grayish yellow	Yellowish gray	Colorless	Light yellow
Sulphuric acid	Grayish yellow- golden-brown	Lemon-pale lemon	E pink-purple; P golden-light yellowish; O distinct	Orange-brown; fine pores distinct
Nitric acid	Grayish lemon-yel- lowish; C swell	No change	PE swell a little, show Str red-brown-deli- cate purple	Lemon-yellow-green- ish yellow; C swell and creep out
Hydrochlo- ric acid	Grayish yellow- lemon-dirty yellow	C swell to double size; PW yellowish; Str distinct	C yellow-colorless; PE golden-delicate greenish yellow	Lemon-dirty yellow; C swell and creep out
Iodine water	Light brown; St black	Dark brown; no starch	Brown; St black	Brown; St black; P yellow
"Lugol's solution"	Brown; St black	Dark brown; no starch	Brown; C and L darker than PW; St black	Brown; St black; P yellow
Ammonia water	No reaction	C swell a little and clear up; granules in the C disappear	Yellow-colorless; C and O¹ Str dis- tinct; P purple	Light yellow; C transparent; Str distinct
Sodium hydroxide	Yellowish, transparent; C swell	C lemon-pale lemon; Str of PW very dis- tinct	Yellow-colorless; P and I yellow; E bright red-brown, distinct	Golden; I red-brown, distinct
Potassium hydroxide	Yellowish, transparent; C swell	C swell threefold, creep out; PW lemon	Same as above	Greenish yellow-yellow-golden; I light red-brown
"Acid Nigrosine"	Parts below the O dark violet	L stain dark violet	L dark violet; later C violet, P light violet	L dark violet

<sup>\*</sup> Abbreviations used in the Synoptical Table:
Shape.—e, elliptical; p, polyhedral; s, spherical; t, tetrahedral.
Pores.—O, large pores; o, small pores; L, lid of a pore.
The number of the pores is expressed by an index figure, thus—O<sup>3</sup>L, three large pores with lids; O<sup>1</sup>, one large pore without a lid; O<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, numerous large pores; o<sup>n</sup>, ous small pores.

Other abbreviations.—C, contents; E, extine; I, intine; P, perine; PW, pollen wall; S, surface; St, starch; Str, structure.

Plant	Amaranthus retro- flexus	Ambrosia artemisiae- folia	Ambrosia trifida	Anthoxanthum odora- tum
Pollinating	End July-first half Aug.	AugOct.	AugSept.	May
Size	23–25 μ	17-22 μ	35-42 μ	$32  imes 35 – 34  imes 37~\mu$
Shape and marks	s on L; S smooth; C coarse	s O <sup>3</sup> L; S obtuse spines equatorially ar- ranged	s O <sup>2</sup> ; S spiculated	e O¹ L; S smooth; C coarse
Color	Grayish yellow	Yellow	Golden	Yellowish
Sulphuric acid	Lemon-orange-light brown-purple; o distinct	Lemon-light yellow- colorless	Yellow-colorless; PE swell, layers distinct; P at last greenish yellow	Lemon-yellow-light brown; grains swell a little
Nitric acid	Lenion; I very deli- cate purple	Greenish yellow-light yellow; C swell and creep out	Colorless; P light yel- lowish; C swell and creep out	Colorless; C swell a little; I lavender and purple; P distinct
Hydrochlo- ric acid	P red-brown-purple	Lemon-golden; C swell	Lemon-deep yellow; C swell and creep out	Yellowish-colorless; PW lemon-bright orange; OL distinct
Iodine water	Light brown; St black	Greenish yellow- brown; no starch	Brown; St black	Light brown; St black; PW dark brown
"Lugol's solution"	Brown; St black; P	Light brown; no starch	Light brown; St black	As in iodine water
Ammonia water	Light brown	Yellowish green; C swell and creep out	Lemon; C shrink	PW lavender
Sodium hydroxide	Light brown; I red- brown	Golden-greenish yel- low; C swell and creep out	Greenish yellow-light yellow; C swell and creep out	PW bright orange very distinct
Potassium hydroxide	Light brown; I red- brown	Golden-greenish yel- low	Lemon; C swell and creep out	As in sodium hydrox- ide
"Acid Ni- grosine"	o dark violet; C later violet	L dark violet	O dark violet	L stains dark violet
Plant	Artemisia absinthium	Aster novae Angliae	Betula populifolia	Carya alba
Pollinating	June-Aug.	AugOct.	FebMar.	May
Size	20-28 μ	24-28 μ	27-32 μ	$38  imes 49$ – $63  imes 70  ext{ }\mu$
Shape and marks	s O <sup>3</sup> L; S spiny	s O <sup>3</sup> L; S spiny; oil drops	s O3 L	e O <sup>3</sup> L; S smooth; C coarse
Color	Yellow	Yellow	Yellow	Light grayish yellow
Sulphuric acid	Dirty yellow-gray- ish blue; PW dirty green, finally light green; C blue gran- ules and crystals	Green-greenish yellow-blue; oil drops blue	Lemon-orange-gold- en-brown	Orange-lemon-light yellow
Nitric acid	Light yellow-color- less	Almost colorless; C swell and creep out, forming drops on the pores	Lemon-greenish yellow; C swell; P and O distinct	Lemon-light yellow; P lemon; E very deli- cate pinkish; C swell and creep out
Hydrochlo- ric acid	Entirely decolorized	C swell and creep out; oil drops deep golden	Yellowish green; C swell and creep out	Golden-light yellow; P light brown

Plant	Artemisia absinthium	Aster novae Angliae	Betula populifolia	Carya alba
Iodine water	Light brown; L yellow; no starch	Light brown; no starch; oil drops greenish	Black (stuffed with starch); P brown; O yellow	Golden-brown; St black
"Lugol's solution"	Brown; L yellow; no starch	Brown; no starch; oil drops greenish	C black (starch); O light brown	Brown; St black; P
Ammonia water	Lemon-light yellow- dirty yellow; C swell	Greenish yellow; C swell a little	Greenish yellow	Yellow; P darker, swells to double size
Sodium hydroxide	Lemon-golden-col- orless	Pale yellow-light yel- low	Golden; Str distinct	Yellow; P golden; O and layers distinct
Potassium hydroxide	As in sodium hy- droxide; oil drops golden	Greenish yellow-light yellow; C swell, be- come distinct	Yellow; Str distinct	As in sodium hydrox- ide
"Acid Nigrosine"	L stain dark violet	L stain dark violet	L stain dark violet	L stain dark violet
Plant	Chenopodium album	Cheno podium ambros- ioides	Chrysanthemum leu- canthemum	Corylus americana
Pollinating	July-Sept.	AugOct.	July-Aug.	FebMar.
Size	20-32 μ	24-28 μ	23-28 µ	28-35 μ
Shape and marks	s on; S smooth; C coarse	p on; S smooth; C coarse	t O <sup>3</sup> L; S spiny; C granulated	t O <sup>3</sup> L; S smooth; C granulated
Color	Dirty yellow	Light yellowish gray	Yellowish gray	Yellow
Sulphuric acid	Orange-bright red- brown; C pale yel- low; PW bright red-brown	Orange-red-brown- ruby-colored-purple	Lemon-greenish yel- low-dirty yellow- colorless	Grayish yellow- orange-light brown
Nitric acid	Bright yellow-pale yellow; C swell a little and creep out	Lemon; o distinct	Colorless	No change
Hydrochlo- ric acid	Yellow	C yellow; I red-brown	Golden-grayish yel- low-colorless; S golden oil drops	Yellowish-greenish; C swell and creep out
Iodine water	Brown; St black	Light brown; St black	C yellow; PW brown; no starch	Brown; St black; P
"Lugol's solution"	Dark brown; St black	Light brown; St black	C dark yellow; PW brown; no starch	Golden brown; St black; O golden
Ammonia water	Lemon-yellow-light yellow	Grains shrink; I bright red-brown	Yellow-golden- lemon; C swell and creep out	Golden-yellow
Sodium hydroxide	Golden-light brown (bright)	Lemon-light brown; o distinct	Lemon; C swell and creep out	Lemon-yellow
Potassium hydroxide	Colden-lemon-yel- low	Lemon; P bright red- brown; grains swell; o distinct	As in sodium hydrox- ide	As in sodium hydrox-ide
"Acid Nigrosine"	o stain at first, later the C	C stain violet very rapidly	L stain dark violet	L stain dark violet

Plant	Cynodon Dactylon	Dactylis glomerata	Dahlia variabilis	Erigeron canadensis
Pollinating	June-Sept.	May-June	SeptOct.	AugSept.
Size	30-38 µ	24 × 28-32 × 39 µ	28-35 μ	16-22 μ
Shape and marks	s O <sup>1</sup> L; S smooth; C granulated	e O¹ L on; S smooth; C coarse	s O <sup>12-20</sup> L; S sharp spines; oil drops	s O <sup>2</sup> L; S spiny
Color	Dirty yellow	Grayish	Yellow	Yellowish gray
Sulphuric acid	Orange-light brown- yellow	Yellowish-pinkish- purple	Orange-light brown- colorless	Light pinkish-yellow
Nitric acid	Yellow-light brown; C swell a little	Yellowish-colorless; C swell and creep out	Greenish yellow-light yellow-colorless; C swell a little	Light yellow-colorless
Hydrochlo- ric acid	Light yellow-color- less	Yellowish; E greenish yellow; I bright red- brown; O greenish yellow	Golden-yellow-light brown; S oil drops	Light yellow-colorless
Iodine water	Brown; St black	Yellow-golden; St black	C brown; PW brown; St black	Light brown; no starch
"Lugol's solution"	As in Iodine water	As in Iodine water	As in Iodine water	As in Iodine water
Ammonia water	Light yellow	Colorless; C swell; P violet-purple	Light brown	Greenish yellow-yel- low
Sodium hydroxide	Yellow; grains swell; O swells, appears very distinct; PW orange	C and E yellowish, swell; P bright red- brown; O distinct	Bright red-orange- light brown-yellow; P swells	Lemon-greenish yel- low-colorless
Potassium hydroxide	As in sodium hy- droxide; O does not swell	As in sodium hydrox- ide	As in sodium hydrox- ide	Lemon-greenish yel- low
"Acid Nigrosine"	L stains dark violet	L stains at first, later the P	L stain dark violet	L stain at first slowly, later the C
Plant	Festuca elatior	Fraxinus americana	Gleditschia triacanthos	Helianthus annuus
Pollinating	June-July	Apr.	May	July-Sept.
Size	23-30 µ	21  imes 25– $24  imes 32  imes$	$28  imes 32$ – $42  imes 46  ext{ }\mu$	30-40 μ
Shape and marks	s O¹ L; S smooth; C coarse	e O <sup>3-4</sup> ; S smooth; C granulated	e O <sup>3</sup> L; S fine-granu- lated	s O <sup>3</sup> L; S sharp- pointed spines; oil drops
Color	Grayish	Brown	Light greenish yellow	Dirty yellow
Sulphuric acid	Colorless; E pinkish- purple	Greenish yellow; O distinct	Golden-colorless; P yellow; E pinkish; O distinct	Lemon-greenish yel- low; oil drops green- blue

Plant	Festuca elatior	Fraxinus americana	Gleditschia triacanthos	Helianthus annuus	
Nitric acid	Colorless; C and PW swell; E dark blue; I cherry-colored	Yellow; P light brown; C swell and creep out	Greenish yellow; C swell three or four- fold	Light (pale) yellow; C swell a little	
Hydrochlo- ric acid	PW swells to double; P bright red- brown; I lemon- colored	1	Greenish yellow; C swell and creep out; P weak greenish yel- low	Yellow (golden)-light (pale) yellow; grains swell and burst	
Iodine water	C light brown; St	Brown; St black	Yellow-chocolate colored; no starch; surroundings of pores not stained	Light brown; no starch	
"Lugol's solution"	As in Iodine water	As in Iodine water	As in Iodine water	As in Iodine water	
Ammonia water	Colorless; PW bright red-brown and lav- ender; O distinct	Greenish yellow; fine pores distinct	Greenish yellow-dirty yellow; P colorless	Lemon-colorless; grains swell	
Sodium hydroxide	Yellowish; PW swells; P bright red-brown	Greenish golden; fine pores distinct	Lemon; P colorless	Bright lemon; L swell	
Potassium hydroxide	C swell; P bright red-brown	Golden-light brown; C swell; fine pores distinct	Greenish yellow- light yellow; C swell and creep out	Lemon-colorless	
"Acid Nigrosine"	L stain dark violet, later the C	Parts of C below the O stain dark violet	L swell and stain dark violet	L swell and stain dark violet	
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Plant	Iva ciliata	Iva xanthifolia	Ligustrum vulgare	Liquidambar st <b>yraciflua</b>	
Pollinating	AugOct.	July-Sept.	May-July	Apr.	
Size	24-28 μ	$14\times1717\times21\mu$	24-35 μ	35–42 μ	
Shape and marks	s O <sup>3</sup> L; S spiny	e O <sup>3</sup> ; S spiny	p O³; S smooth; C granulated	s on L; S smooth	
Color	Grayish yellow	Grayish yellow	Yellow	Yellowish	
Sulphuric acid	Lemon-light yellow	Lemon-light yellow; Str of PW distinct	Orange-red-brown- brown	Orange-light brown; P golden	
Nitric acid	Light brown; C swell and creep out	Lemon; C swell and creep out	Greenish yellow-dirty yellow; C swell to double size and creep out	Yellow; grains swell; P brown glimmer; o distinct	
Hydrochlo- ric acid	Light yellow	Light yellow-color- less; P light pinkish	Light greenish yellow; C swell and creep out	Yellowish-light brown; P light brown	
Iodine water	Light yellow; St black	Light brown, later dark brown; St black	Light brown; no St traceable	Yellow; St black	
"Lugol's solution"	As in Iodine water	As in Iodine water	As in Iodine water Golden-brown;		

Plant	Plant Iva ciliata Iva xanthifolia L		Ligustrum vulgare	Liquidambar styraciflua
Ammonia water	Lemon-light yellow- colorless; Str dis- tinct	Yellow-colorless; grains swell; Str dis- tinct	Greenish yellow-dirty yellow	Yellow; grains swell a little; o distinct
Sodium hydroxide	Lemon-greenish lemon; grains swell a little	Greenish yellow-light yellow-colorless	Greenish brown- greenish yellow	Light yellowish-color- less; grains swell
Potassium hydroxide	As in sodium hydroxide	Greenish yellow-colorless; Str distinct	Greenish brown- greenish yellow; P colorless; C swell	Yellow-greenish yel- low; grains swell a little
"Acid Nigrosine"	L stain first, later C	Three dark violet cone-shaped stop- pers from the O to the centre	Places below the O stain dark violet	Whole grains stain dark violet
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Plant	Lolium perenne	Medicago sativa	Melilotus alba	Morus alba
Pollinating	May-June	May-Oct.	May-Oct.	Apr.
Sise	$24 \times 31  32 \times 39 \; \mu$	31 × 35-42 × 45 µ	20-28 µ	17-21 μ
Shape and marks	e O <sup>1</sup> L; S smooth; C granulated	e O <sup>3</sup> L; S small reticu- lations; C granu- lated	t O <sup>2</sup> L; S cone-shaped projections from pores; C granulated	s O <sup>2</sup> L; S smooth; C coarse
Color	Yellowish	Grayish yellow	Greenish yellow	Light gray
Sulphuric acid	Light yellow-light brown; O distinct	Pinkish yellow-light brown-grayish; PW yellow	Yellow-light lemon- light yellow	Light brown
Nitric acid	Lemon-violet-pur- ple; P swells	Light brown; C swell; grains burst; PW yellow	Light yellow	P colorless; E pinkish- purple
Hydrochlo- ric acid	Grains swell and burst	Colorless; C swell to double size and creep out	Colorless	C swell and creep out; E light pinkish
Iodine water	Yellow; no St	Brown; no St	Brown; PW yellow;	Yellow-brown; St black
"Lugol's solution"	Yellow; P bright red- brown	As in Iodine water	As in Iodine water	Brown; P light brown; St black
Ammonia water	No change	Light yellow; O dis- tinct; grains swell	Lemon-light yellow drops in the C	I pinkish-purple
Sodium hydroxide	Yellow; P bright red- brown; granules very distinct	Greenish yellow-dirty yellow	Lemon-colorless; C swell to double size and creep out	Yellow-colorless; C swell; Str distinct
Potassium hydroxide	Yellow; P red-brown, later light brown; granules distinct	Pale yellow; grains swell threefold and burst; PW choco- late-colored	As in sodium hydrox- ide	As in sodium hydrox- ide
"Acid Nigrosine"	L stains dark violet	L stains dark violet	L stains dark violet	At first the L of the O stain; later the P

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Plant	Panicum anceps	Phleum pratense	Pinus austriaca	Plantago lanceolata
Pollinating	July-Sept.	June-Aug.	May	May-Sept.
Size	30–38 µ	25 × 31-33 × 35 μ	$31 \times 50-50 \times 70 \mu$ ; without air-sacs, $31 \times 40-38 \times 55 \mu$	20 × 28 μ
Shape and marks	s O¹ L; S smooth; C granulated	e O¹ L; S smooth; C granulated	e O <sup>2</sup> L? S two air-sacs; C granulated	s O <sup>12</sup> L; S smooth; C coarse
Color	Yellowish gray	Yellow	Light yellow	Yellowish gray
Sulphuric acid	Light brown-orange- golden; PW golden	Light yellow	Orange-flesh-red; P yellow; air-sacs later flesh-red	Orange-pinkish-color- less; PE golden
Nitric acid	Colorless; C clear up	Yellowish-colorless; grains swell to double size	Yellow; P lemon	Golden-yellow; P colorless; E red- brown; L swells
Hydrochlo- ric acid	C colorless	No color reaction; C swell and creep out	C light dirty brown; grains swell to double size	Light red-brown-light brown-colorless; E red-brown; C swell
Iodine water	C dark brown; PW red-brown; St black	Yellow-golden; St black	Lemon-greenish yellow; St black	Golden; St black
"Lugol's solution"	As in Iodine water	Brown; St black	Light brown; St black; grains swell a little	Greenish yellow; St black
Ammonia water	No change	No change	Lemon-dirty yellow	Greenish yellow-color- less; P bright red- brown; O distinct
Sodium hydroxide	Yellow-pale yellow	Light yellow; Str dis- tinct	Yellow; the air-sacs show the color re- action later	Grayish yellow; P red- brown; L swell
Potassium hydroxide	Yellow	No color reaction; O distinct	As in sodium hydrox- ide, but the color greenish yellow	As in sodium hydrox- ide
"Acid Nigrosine"	L dark violet	At first the L stains dark violet, later the whole grain	No distinct reaction; after a time the air- sacs stain	L stains dark violet
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Plant	Plantago major	Platanus occidentalis	Poa pratensis	Polygonum persicaria
Pollinating	June-Sept.	Мау	May-Sept.	AugSept.
Size	20-25 μ	20-34 μ	31-37 μ	28-53 μ
Shape and marks	s O <sup>6</sup> L; S smooth; C coarse	t O <sup>3</sup> ; S smooth; C coarse	8 O <sup>1</sup> L; S smooth; C coarse	s On L; S hexagonal reticulations; C granulated
Color	Light yellowish	Dirty yellow	Greenish yellow	Light yellow
Sulphuric acid	Lemon-colorless	Lemon-light brown; PW yellow	Orange-yellowish- brown	Greenish yellow- lemon-light brown; surface markings dis- tinct
Nitric acid	Yellow (weak)-colorless	Light brown; C swell	Light yellow; E bright red-brown; grains swell	No color reaction, C swell and creep out; reticulations very distinct
Hydrochlo- ric acid	Colorless; grains burst	Lemon; no change	Light brown; P and E bright red-brown	No color reaction; Str distinct

Plant	Plantago major	Platanus occidentalis	Poa pratensis	Polygonum persicaria
Iodine water	Dark brown; O col- orless; St black	Brown; very fine and small starch grains which stain black	Light brown; no St	Light brown-greenish yellow; St black
"Lugol's solution"	As in Iodine water	As in Iodine water	Brown; no St	As in Iodine water
Ammonia water	No change	Lemon-dirty yellow; C swell a little	Light yellow; P bright red-brown	Greenish yellow
Sodlum hydroxide	Light yellow	Pale yellow-colorless	Light yellow; P and E bright red-brown	Greenish yellow
Potassium hydroxide	Colorless	Lemon; C swell and creep out	As in sodium hydrox- ide	Greenish yellow-dirty yellow
"Acid Nigrosine"	L stains dark violet, later the C	Places below the O stain dark violet; later C stain	L stains dark violet; later C stain	L stain dark violet
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Plant	Populus balsamifera	Pyrus Malus	Quercus alba	Quercus coccinea
Pollinating	Mar.	May	May	May
Size	28-40 μ	34-35 μ	28-34 μ	24-35 μ
Shape and marks	s on; S fine-granu- lated	t O <sup>3</sup> ; S cone-shaped projections from pores; C coarse	s O <sup>3</sup> L; S cone-shaped projections from pores	tO <sup>3</sup> L; S cone-shaped projections from pores; C coarse
Color	Yellow	Yellowish gray	Yellowish	Dirty yellow
Sulphuric acid	Yellowish green- pinkish; P bright yellow	Yellow-lemon-dirty yellow; PW lemon	Orange-red-brown; C yellow	Red-brown-light Indian red
Nitric acid	No reaction	Light yellow	Lemon-light yellow- colorless; C swell threefold and creep out	Lemon-light yellow- colorless; C swell threefold and creep out
Hydrochlo- ric acid	Greenish yellow-colorless, transparent	Colorless; C swell to double size and creep out	Yellow-lemon-green- ish yellow	Light yellow; PW darker; O colorless; C swell a little
Iodine water	C brown; St black; I red-brown	Brown; St black	C brown; PW yellow; very few small starch grains, which stain black	Brown; no starch
"Lugol's solution"	As in Iodine water	As in Iodine water	As in Iodine water	As in Iodine water
Ammonia water	PW swells and shows the layers	Colorless	Greenish lemon- greenish yellow	Yellow-lemon-dirty yellow
Sodium hydroxide	No change	Lemon-yellowish; C swell a little	Golden; PW light brown; O colorless	Lemon-dark greenish yellow; PW light brown; O colorless
Potassium hydroxide	Light yellow; grains burst	Lemon-yellowish-col- orless	Golden-lemon-yel- low; C swell a little; globoids distinct	Lemon-light yellow; C swell a little
"Acid Nigrosine"	C dark violet; PW light violet	Places below the O stain dark violet	L stain dark violet	L stain dark violet, later the places be- low the O; projec- tions light violet

Plant	Quercus rubra	Robinia pseudacacia	Rudbeckia laciniata	Rumex acetosella
Pollinating	May	AprMay	AugSept.	May-Aug.
Size	24-35 μ	28-41 μ	20-25 μ	21 × 24-24 × 28 µ
Shape and marks	s O <sup>3</sup> L; S smooth; C coarse	t O <sup>3</sup> L; S fine-granu- lated	s O <sup>3</sup> L; S spiny	e O4; S smooth; oil drops; C coarse
Color	Dirty yellow	Grayish yellow	Yellow	Yellow
Sulphuric acid	Red-brown-light brown	Golden-colorless; P yellow; E light pink- ish	Lemon-greenish lemon; PW bluish green	Orange-light purple
Nitric acid	Yellow-light yellow; grains swell and burst	C swell four or five- fold and creep out	Colorless; grains swell a little	Colorless; C swell a little and part creeps out
Hydrochlo- ric acid	Light yellow; C swell	No color reaction; C swell and creep out	Light yellow; Str dis- tinct	Colorless; P yellowish
Iodine water	Light brown; no starch	Light brown-dark brown; St black	Brown; no starch	C yellow; P light brown; St black
"Lugol's solution"	As in Iodine water	Light brown-red- brown; very small starch grains stain black	As in Iodine water	C and P light brown St black
Ammonia water	Lemon-greenish yel- low-dirty yellow	No change	Light yellow-color- less	Grains shrink; Str of P and O distinct
Sodium hydroxide	Golden	Yellow; grains swell and burst	Lemon-greenish yel- low-dirty yellow	Yellowish-colorless; & warty
Potassium hydroxide	Golden	Yellow; C swell and creep out	As in sodium hydrox- ide	Grains swell; S warty
"Acid Nigrosine"	O stain dark violet, later the places be- neath the O	O stain dark violet, later the places be- neath the O	L stain dark violet	O stain light violet very slowly
Plant	Solidago canadensis	Taraxacum officinale	Trifolium pratense	Typha latifolia
Pollinating	AugOct.	All year	AprNov.	June
Size	15 × 21-18 × 24 μ	35-50 μ	28-41 μ	$35  imes 42 – 38  imes 45  ext{ }\mu$
Shape and marks	e O <sup>3</sup> L; S obtuse spines; oil drops	s O <sup>12-20</sup> L; S short blunt spines; oil drops	s O <sup>3</sup> L; S cone-shaped projections from pores	s O <sup>1</sup> L; pollen grains always in aggrega- tions; S smooth
Color	Yellowish	Golden	Grayish	Yellow
Sulphuric acid	Greenish yellow-very light brown	PW purple; oil drops blue	Yellowish	Orange-light brown- light yellow; P light yellow; C light pink- ish
Nitric scid	Colorless; C swell and creep out	Light yellow-color- less; C swell and creep out	Light yellow; C swell and creep out	Yellow; C swell and creep out
Hydrochlo- ric acid	Lemon-dirty yellow	C swell; grains burst	C swell and creep out; no color reaction	As in nitric acid

Plant	Solidago canadensis	Taraxacum officinale	Trifolium pratense	Typha latifolia
Iodine water	Brown; no starch	Brown; no starch traceable; oil drops bluish green	C brown; no starch traceable	Golden; St black
"Lugol's solution"	As in Iodine water	As in Iodine water	As in Iodine water	As in Iodine water
Ammonia water	Colorless	Pale yellow; grains burst	No change	Golden
Sodium hydroxide	Greenish yellow; Str distinct	Lemon-dirty yellow	Yellowish; C swell and creep out	Golden-light yellow; Str of PW distinct
Potassium hydroxide	Light yellow; Str distinct	Pale yellow	C swell and creep out	As in sodium hydrox ide
"Acid Nigrosine"	L stain dark violet	After a long time the O take the color	L dark violet; projec- tions light violet	L stains dark violet
Plant	Ulmus americana	Xanthium spinosum	Zea Mays	
Pollinating	MarApr.	AugSept.	June-July	
Sise	25-35 μ	22-28 μ	$70  imes 75$ – $85  imes 88  ext{ }\mu$	
Shape and marks	p O <sup>5</sup> L; S smooth; C fine-granulated	s O <sup>3</sup> L; S smooth; C coarse	e O <sup>1</sup> L; S smooth; C granulated	
Color	Greenish yellow	Light brown	Light yellowish	
Sulphuric scid	Orange-light yellow- ish-colorless; PE distinct (Str)	Greenish yellow- brown	Yellow-golden-red- brown; surroundings of the O light yellow	
Nitric acid	Lemon-greenish yel- low-colorless; Str of PW very distinct	Light yellow	Light yellow; C swell; O distinct	
Hydrochlo- ric acid	Orange-yellow	Greenish yellow; Str of PW distinct	Yellow-lemon-gray- ish yellow; C swell; O golden	
Iodine water	Weak yellowish; no starch	Light brown; no starch	C light brown; St black; PW bright red-brown	
"Lugol's solution"	Dark brown, no starch; PE light brown; O distinct	Brown; no starch	As in Iodine water	
Ammonia water	Golden-greenish yel- low; O and PW dis- tinct	No change	Grayish yellow; grains swell; O dis tinct	
Sodium hydroxide	Golden; C swell	Lemon; I red-brown; Str of PW distinct	Yellow-lemon	
Potassium hydroxide	Golden	As in sodium hydrox- ide	Yellow-lemon; C coarse; P red-brown O lemon	;
"Acid Nigrosine"	L stain dark violet	O and parts of C be- low stain dark violet	O stains dark violet; PW stains later	

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# DETERMINATION OF TOTAL NITROGEN, NITRATE-NITROGEN, AND TOTAL NITROGEN NOT INCLUDING NITRATE-NITROGEN: FURTHER OBSERVATIONS ON A MODIFICATION OF THE OFFICIAL SALICYLIC-THIOSUL-PHATE METHOD<sup>2</sup>

#### EMERY R. RANKER

Associate Physiologist, Office of Cereal Investigations, Bureau of Plant Industry,
United States Department of Agriculture
Formerly Rufus J. Lackland Research Fellow in the Henry Shaw School of Botany
of Washington University

## INTRODUCTION

The data here presented are in continuation of a series of investigations previously reported (Ranker, '25). These previous data were presented in support of a proposed modification of the official salicylic-thiosulphate method for the determination of total nitrogen including nitrate-nitrogen. The recommended procedure is as follows:

"Place the sample in an 800-cc. Kjeldahl flask; adjust to neutrality or make slightly alkaline; if water is present evaporate just to dryness on a water bath under vacuum. Add 35-40 cc. of salicylic acid mixture (1.0 gm. of salicylic acid to 30 cc. of concentrated nitrogenfree sulphuric acid); mix thoroughly and allow to stand for at least an hour with occasional shaking (if organic matter is present, stopper tightly with a rubber cork and allow to stand over night). Add 5 gms. of sodium thiosulphate and heat for 5 minutes with a low flame; cool; add 7-10 gms. of anhydrous sodium sulphate and a pinch of copper sulphate. Digest for an hour at the boiling point after the solution clears; just before the solution solidifies dilute to an estimated volume of 400 cc.; cool completely. Add a small piece of paraffin, 100 cc. of a saturated solution of sodium hydroxide, and a piece of

<sup>1</sup> By the unqualified term "total nitrogen" is meant the sum total of all forms of nitrogen present in the particular sample of material being analyzed.

<sup>2</sup> The second part of a series of investigations carried out at the Missouri Botanical Garden in the Graduate Laboratory of the Henry Shaw School of Botany of Washington University, and submitted as a thesis in partial fulfillment of the requirements for the degree of doctor of philosophy in the Henry Shaw School of Botany of Washington University.

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mossy zinc; connect immediately to the distillation apparatus and distill 150-200 cc. over into standard acid during a period of 1 hour. Titrate the standard acid to neutrality with standard alkali and calculate the amount of nitrogen present."

In the interest of brevity this proposed method will be referred to as the "modified official method." Certain further applications of the modified official method seemed desirable, and at the same time such an extension would afford opportunity to test its accuracy more extensively. The work has progressed along several lines and certain phases of the investigation are being continued.

In the work previously referred to (Ranker, '25, table 1) it was found that only  $92.7 \pm .90$  per cent of the total nitrogen present was recovered by the modified official method, from samples containing "50 mgs. nitrate-nitrogen plus 0.5 gm. In contrast  $98.1 \pm .64$  and  $99.5 \pm .00$  per cent of sucrose." the total nitrogen were recovered by the comparison method used. Identical samples were used in all cases. From these data it was concluded (page 371) that "if sugar is present in abundance a slight loss of nitrate-nitrogen may occur, due to the reducing action of the sugar." It was concluded further that "this loss would be very slight in actual practice since the nitrate-nitrogen content of plants is small." The last conclusion is based on assumption only. In order to ascertain the true condition that would maintain "in actual practice" some analyses were made to determine the accuracy of the modified official method for samples of plant materials high in sugar and nitrate-nitrogen content. These data are given in table 1.

Strowd ('20) concluded that "the determination of nitrates in plants by finding the difference between the Kjeldahl-Gunning-Arnold method and the Kjeldahl method modified to include nitrates is unsatisfactory." Other workers have noted this difficulty also. Strowd explained this discrepancy on the possible basis that "appreciable amounts of nitrate were apparently reduced without zinc and salicylic acid." However, no data were presented in support of this conclusion (Strowd, '20). In connection with this phase of the problem at least 3 conditions must be recognized:

(1) The inaccuracy of the official salicylic-thiosulphate method,

occasioned by the presence of water in the sample (Ranker, '25), has made impossible a critical comparison with other methods for the determination of nitrate-nitrogen by difference.

- (2) The accuracy of the Kjeldahl-Gunning-Arnold method for the determination of "total nitrogen not including nitratenitrogen" only, is generally accepted. In the presence of nitratenitrogen, however, this method may not be accurate. Such a possibility must be recognized since nitrates are quite universally present in plants; such forms as Selaginella and celery, for example, have a high nitrate content.
- (3) The Devarda method is frequently used as a comparison method to test the accuracy of other methods for the determination of nitrate-nitrogen only, in plant materials. Allen ('15) and Davisson ('18) call attention to the possibility that some organic-nitrogen substances may be acted upon in the process. Such a condition would impair the value of the Devarda method for such comparisons and for unqualified determinations of "nitrate-nitrogen" in plant materials.

The above phases of the problem of nitrogen determination were investigated by making certain analyses, as follows:

- 1—To determine the accuracy of the Kjeldahl-Gunning-Arnold method for the determination of "total nitrogen not including nitrate-nitrogen" only, in nitrate-free samples to which a known amount of nitrate-nitrogen is added.
- 2—To determine whether or not nitrate-nitrogen only can be quantitatively determined by finding the difference between the modified official method (Ranker, '25) and the Kjeldahl-Gunning-Arnold method.
- 3—To continue with the study of the influence of certain details of manipulation upon the accuracy of the modified official method in particular.

# MATERIALS

The various determinations and analyses were made of samples taken from the various soil and plant materials listed below. Most of these materials are on the plant side and most of them were in solution when used. At the time of sampling, the solutions were free from any precipitate or suspended matter; if not,

they were filtered before samples were taken. All comparable samples were measured into the Kjeldahl flasks (800 cc.) with the same pipette, at the same time and temperature, and all other conditions of making comparable samples were as nearly identical as possible. The numbers assigned to the various materials correspond to the numbers used in the tables and in the text, as indicating the stock materials from which the samples were taken. Though 2 sets of samples may bear the same number they are not necessarily comparable unless they appear in the same table; they may have been measured out at different times and temperatures. The materials analyzed were as follows:

1—Aspergillus niger. 2—Fusarium culmorum.

Whole cultures, including residual solutions, of the above organisms were cultured under sterile conditions, in 100-cc. flasks containing exactly 25 cc. Pfeffer's nutrient solution to which 1 per cent glucose had been added. The cultures were grown until a vigorous heavy mat had formed. The entire contents of 1 culture flask constituted 1 sample, being transferred without loss to a Kjeldahl flask for determination of nitrogen.

3—Aspergillus niger.

5-Phoma Betae.

4—Fusarium culmorum.

Residual solutions only of the above organisms were cultured, under sterile conditions, in 300-cc. flasks containing 100 cc. of nutrient solution (refer to No. 1 and 2). At time of sampling the cultures were boiled for 5 minutes and filtered. Analyses were made of 25 cc. of the clear filtrate per sample.

6—Tobacco leaves (water extract).

7—Geranium leaves (water extract).

8—Pea leaves and terminals (water extract).

9—Celery leaves and stalks (water extract).

10—Algae mixture, mostly Spirogyra

(water extract).

11—Tomato fruits, ripe (water extract).

12—Tobacco leaves and tops (alcohol extract).

13—Geranium leaves (alcohol extract).

14—Pea leaves and terminals (alcohol extract).

15—Celery leaves and stalks (alcohol extract)

16—Tomato fruits, ripe (alcohol extract).

The fresh material of the above plants was ground through a food-chopper; any drippings produced were added; 1 volume of water or 1 volume of 70 per cent alcohol was added; the mixture was boiled (refluxed in the case of alcohol) for 20-30 minutes and filtered hot; 25-cc. samples were taken from the filtrate for analyses.

17—Greenhouse soil plus mushroom compost (water extract).

One kgm. dry material plus 1 L. H<sub>2</sub>O; stirred rapidly 4 hrs.; allowe to precipitate; decanted; filtered; 25-cc. samples taken from filtrat

18—Sugar-cane.

20—Pea leaves and terminals.

19—Sugar-beet.

Water extracts of these materials were prepared in the same manner as indicated for Nos. 6-16.

- 21—Greenhouse soil containing mushroom compost (water extract)
  Prepared from same materials and in same manner as No. 17.
- 22—Mushroom compost only (cold water extract). Prepared same as No. 17.
- 23—Mushroom compost (autoclave extraction).

Prepared same as No. 17 except the mixture was autoclaved, not stirred.

24-Mushroom compost, KNO<sub>3</sub>.

Twenty-five cc. No. 22 and 74 each, per sample.

25—Aspergillus niger, KNO<sub>3</sub>.

Twenty-five cc. No. 3 and 74 each, per sample.

26-Fusarium culmorum, KNO3.

Twenty-five cc. No. 4 and 74 each, per sample.

27—Crude peat (water extract).

30—Selaginella apus (expressed sap).

28—Sphagnum moss (dry) (water extract).

31—Selaginella apus (alcohol extract).

29—Selaginella apus (water extract).

32—Selaginella apus (water extract).

Prepared same as Nos. 6-16.

Numbers 33-50 refer to various samples of solution cultures of barley, wheat, and peas. These materials were used for qualitative analyses only; the nature and preparation of the samples are indicated in the various tables where the data of their analyses are recorded.

52-Alanin, KNO<sub>3</sub> solution.

Twenty-five cc. alanin solution (1.277 gm. per liter) and No. 74 each, per sample.

53-Asparagin, KNO<sub>3</sub> solution.

Twenty-five cc. asparagin soln. (0.873 gm. per liter) and No. 74 each, per sample.

54-Sugar-cane, KNO<sub>3</sub>.

Twenty-five cc. No. 18 and 74 each, per sample.

55-Urea solution.

Two and one-tenths gm. per liter; 25 cc. per sample.

57—Sugar-cane (water extract). Prepared same as No. 18.

58—Sugar-cane residue, pulp.

Residue from hot-water extractions; washed until free from NO<sub>3</sub> and NH<sub>3</sub>.

59—Mushroom compost (water extract). Prepared same as No. 17.

60—Sugar-beet (water extract). Prepared same as Nos. 6-16.

61—Sugar-beet residue, pulp.

Residue from hot-water extractions; washed until free from NO<sub>2</sub> and NH<sub>2</sub>.

62—Sugar-beet (third water extract).

Third wash water from No. 61, free from NO<sub>3</sub> and NH<sub>2</sub>.

64—Urea, KNO<sub>3</sub> solution. Twenty-five cc. No. 55 and 74 each, per sample.

65—Sugar-beet, KNO<sub>3</sub>.

Twenty-five cc. No. 60 and 74 each, per sample.

70-Uric acid solution.

Five gms. per liter; 50 cc. per sample.

71—Taka diastase solution.

Seven and one-half gms. per liter; 50 cc. per sample.

72—Pea seed, germinated.

These seed (Gradus variety) were germinated for 5 days, after which they were mashed in a mortar to a fine pulp in 5 times their weight (dry wt.) of water. This material was free from nitrates; there was a trace of ammonia.

73—Heavy clay-loam soil (water extract). Prepared same as No. 17.

74—KNO<sub>3</sub> solution.

This solution contained 1.443 gm. of KNO<sub>3</sub> per liter; by analysis, 25 cc. contained  $4.79 \pm .005$  mgs. N. In all cases those samples which contained added nitrate-nitrogen received 25 cc. of this solution. The nitrogen content of this solution did not vary throughout the period of these investigations as determined by frequent control analyses of 25-cc. samples.

#### METHODS

The following methods were used for the determination of nitrogen:

- (1) The modified official method as reported by Ranker ('25) for the determination of total nitrogen including nitrate-nitrogen.
- (2) A modification of the Devarda method (Ranker, '25) for the determination of nitrate-nitrogen, and for the determination of total nitrogen including nitrate-nitrogen as a comparison method for the modified official method.
- (3) The Kjeldahl-Gunning-Arnold method (Association of Official Agricultural Chemists, '25, page 8, No. 24) for the determination of total nitrogen not including nitrate-nitrogen.

The acid and alkali used in titration were standardized against benzoic acid obtained from the U. S. Bureau of Standards (Sample No. 39B). Fiftieth normal acid and alkali were used in all titrations and the normality factors were redetermined frequently to avoid possible errors from this source. All titrations were carried to the complete disappearance of any red tinge as the end-point of methyl red. With N/50 alkali methyl red was found to be much more sensitive than cochineal and the end-point which was used is practically identical with that of cochineal.

Every determination reported in this paper was checked, qualitatively, for the loss of nitrogen at every stage in the process of analysis. The methods of conducting these qualitative control determinations may be grouped under 3 headings:

- 1. Qualitative tests made during the process of evaporation of the sample under vacuum: All the vapors evolved in this process were passed through a weak solution of sodium bicarbonate and collected in a second flask, both of which are illustrated in fig. 1. The solution thus collected was tested, (A) for the presence of nitrites and nitrates by the diphenylamine test, and (B) for the presence of ammonia by the use of Nessler's reagent.
- 2. Qualitative tests made following the addition of the salicylic acid mixture to the sample: Any vapors or fumes formed during this process were forced through a weak solution of sodium bicarbonate by blowing gently on the air inlet tube of the device illustrated in fig. 2. In order to obtain as concentrated a solution for the tests as possible, not more than 10–20 cc. of the bicarbonate solution was used as an absorbing solution. The solution thus

obtained was tested, (A) for the presence of nitrites and nitrates by the diphenylamine test, and (B) for the presence of ammonia by the use of Nessler's reagent.

3. Qualitative tests made during the process of acid digestion: The fumes evolved during this process were led through and collected (by the use of a filter pump) in a flask containing approximately 50 cc. of distilled water. The solution thus col-

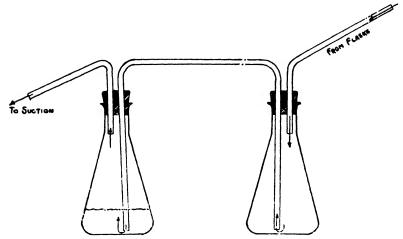


Fig. 1. Arrangement of flasks for the collection of vapors evolved during the evaporation of samples under vacuum and for the collection of fumes evolved during the process of acid digestion.

lected was tested (A) for the presence of nitrites and nitrates by the diphenylamine test and (B) for the presence of ammonia by the use of Nessler's reagent.

The 3 groups of qualitative tests, as indicated above, provide a satisfactory check on the accuracy of each stage of the procedure as recommended in the modified official method. Such qualitative tests are a safeguard against carelessness and should be considered as much a part of quantitative procedure as the actual quantitative determination itself. The data obtained from the qualitative tests are presented as the basis for certain recommendations in regard to details of manipulation and procedure. The above capital letters ("A" and "B") are used throughout the various tables as column headings in the same significance as used above; that is, "A" indicates the results

obtained with the diphenylamine test and "B" indicates results obtained with Nessler's reagent.

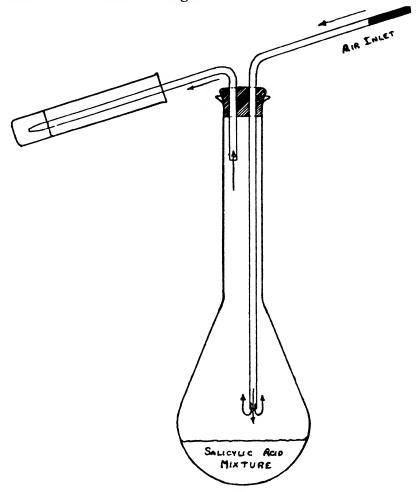


Fig. 2. Method of collecting fumes evolved from samples following the addition of the salicylic acid mixture. The solution collected in the apparatus of figs. 1 and 2 was tested for the presence of nitrites, nitrates, and ammonia.

The evaporation of all samples, which were evaporated, was accomplished on the water bath, under vacuum or as in fig 1.

All probable errors reported in this paper were calculated according to the following formula:

$$\left(E_{\rm m} = \frac{0.6745\sigma}{\sqrt{n}}\right)$$

## EXPERIMENTAL DATA

The prime motive in this investigation has been two-fold: (1) to examine, critically, the accuracy of the modified official method (Ranker, '25), and (2) to extend the application of the method to as many samples of soil and plant materials as time and opportunity would permit. This plan required investigation of the subject from several angles. The data obtained will be presented very much according to the general outline in the introductory remarks.

Before entering into the presentation of data certain terms need definition. The term "vacuum" has reference to an equivalent of not less than 22–23 inches of mercury. By the term "just to dryness" reference is made to that stage in the evaporation process evidenced by the following conditions: the sample has just ceased ebullition and there may or may not be water of condensation in the neck of the flask. By the term evaporation to "ash dryness" the following condition is indicated: the sample, the entire inside of the body of the flask and most of the neck of the flask are dry. Evaporation to "partial dryness" denotes a condition in which some free moisture is present in the sample when the evaporation process is stopped; that is, the sample itself is moist.

In connection with the data of table I certain points should be noted. The probable errors are entirely satisfactory; for example, in the case of sample No. 18 (sugar-cane) the nitrogen content as found in 15 determinations by the modified official method is  $11.5 \pm .006$ . The quantitative results are checked by suitable qualitative tests so that the accuracy of the procedure is definitely known. All of the analyses which were made by these two methods have been included in the table. In practically all of the determinations the accuracy of the modified official method is satisfactory. There are a few cases of rather serious disagreement, however, between the results obtained by the 2 methods. this connection the determinations for sample No. 6 (tobacco) are noticed at once. This discrepancy seems to exist, also, with the alcohol extract of this material (No. 12), in each case the determination by the Devarda method being low. After several tests this discrepancy was determined to be due to the volatile

TABLE I ACCURACY OF THE MODIFIED OFFICIAL METHOD FOR TOTAL NITROGEN IN VARIOUS SAMPLES OF BIOLOGICAL MATERIALS

(arithmetical mean and probable error)

			Analys Devarda		od	Analysis by modified official metho		hod		
				Qu tests	† for			tests†,		
	Sample determined	No. of trials	Nitrogen found (mgs.)	dur diges	ing	Nitrogen found (mgs.)		ring ration	Dur diges	
No.*	Description*		(	A‡	Bţ	(=======	A‡	B‡	A‡	B‡
1	Aspergillus niger (whole cult. incl. soln.)	15	2.2 ± .014	_	_	2.3 ± .009		_	_	
2	Fusarium culmorum (whole cult. incl. soln.)	15	2.3 ± .024	_	_	2.4 ± .010		_	_	_
3	Aspergillus niger (residual solution only)	17	0.84 ± .024	_	_	0.83±.005		_	_	_
4	Fusarium culmorum (residual solution only)	18	1.4 ± .021	_		1.4 ± .013	_		_	_
5	Phoma Betae (residual soln.)	15	2.1 ± .011	_	_	2.1 ± .030		_	_	_
6	Tobacco (water extract)	15	11.4 ± .027	_	_	$12.6 \pm .039$		_	_	_
7	Geranium (water extract)	15	$3.7 \pm .028$	-	_	3.9 ± .020		_	_	-
-8	Garden pea (water extract)	14	$21.6 \pm .045$	-	_	$21.8 \pm .021$		_	_	_
9	Celery (water extract)	15	13.1 ± .030	-	_	13.6 ± .009		_	_	_
10	Algae mixture (water extract)	15	0.9 ± .003	_	_	0.9 ± .005			_	_
11	Tomato fruit (water extract)	15	10.0 ± .018	=	_	10.0 ± .021	_	_	_	_
12	Tobacco (alcohol extract)	15	$5.3 \pm .028$	-	_	5.5 ± .007		_	_	_
13	Geranium (alcohol extract)	15	6.0 ± .039	-	_	6.2 ± .011		_	_	_
14	Garden pea (alcohol extract)	15	9.0 ± .039	-	_	9.7 ± .015	_	_	-	=
15	Celery (alcohol extract)	15	6.1 ± .027	-	_	$6.0 \pm .018$	_	_	_	_
16	Tomato fruit (alc. extract)	15	15.2 ± .009	_		$15.2 \pm .012$	_	_	_	_
17	Greenhouse soil containing mushroom compost (water extract)		3.9 ± .013	_	_	3.9 ± .006	_	_	_	_
18	Sugar-cane (water extract)	15	11.4 ± .045	=	-	11.5 ± .006			_	-
19	Sugar-beet (water extract)	15	10.9 ± .067	-	-	$10.7 \pm .029$	_	_	_	-
20	Garden pea (water extract)	15	$29.3 \pm .053$	-	-	$29.3 \pm .014$		-	1_	1-

<sup>\*</sup>The sample numbers correspond to those numbers in the discussion of materials; the method of preparation and description of the samples, also, are given there.

† The methods of conducting these tests are discussed under "Methods."

‡ The letter "A" is used to designate the diphenylamine test for the presence of nitrites and nitrates; "B," to designate the test with Nessler's reagent for the presence of ammonia.

nature of the nicotine contained in the tobacco material. During the alkaline distillation with Devarda's alloy a large part of the nicotine present is volatilized and driven over into the standard acid sample; some of it is absorbed but much of it passes out into the surrounding atmosphere. Such a loss would contribute toward low results by the Devarda method. This condition is demonstrated by the odor of the nicotine given off and may be determined more or less quantitatively by collecting such vapors in sulphuric acid and analyzing after the Kjeldahl method.

Certain other disagreements exist in the data of table 1; for example, in samples No. 6, 9, 12-14. In this connection attention must be called to a rather outstanding fact; in all cases (except sample No. 6 as noted above) in which disagreements are evidenced the relatively low magnitude of the probable error obtained by the modified official method indicates its greater accuracy and reliability. This fact is demonstrated by a consideration of the data for sample No. 9 (13.1  $\pm$  .03 as compared with 13.6  $\pm$ .009), for sample No. 12 (5.3  $\pm$  .028 as compared with 5.5  $\pm$  .007), and for sample No. 13 (6.0  $\pm$  .039 as compared with 6.2  $\pm$  .011). An examination of other sets of determinations reveals the same evidence. The disagreement existing in the case of sample No. 8 does not appear in the case of sample No. 20, although both samples were taken from similar materials. Taken as a whole the data indicate certain points worthy of mention: there is rather satisfactory agreement between the methods; many of the determinations are identical; the data are based on a sufficiently large number of determinations; and the superior accuracy of the modified official method is indicated. Satisfactory results were obtained in the analysis of samples high in sugar content; that is, samples having a high reducing power (samples No. 18 and 19 in table I; samples No. 54 and 60 in table II).

The accuracy of the modified official method is shown, further, by the data of table II. The qualitative results given are considered to be of more value as indicators of the accuracy of the method than purely quantitative data only. The possible errors of distillation and titration are eliminated, thus allowing the distinctive processes of the method (evaporation under

vacuum, addition of the acid mixture, and digestion) to be checked under more exacting conditions. The procedure followed was exactly that previously recommended (Ranker, '25) except that certain of the samples were not adjusted to neutrality (this phase of the subject will be considered in connection with the data of tables v and vi).

TABLE II

ACCURACY OF THE MODIFIED OFFICIAL METHOD FOR TOTAL NITROGEN
AS INDICATED BY QUALITATIVE TESTS ON THE PROCEDURE
RECOMMENDED\*

		Qualitative tests made†					
	Sample analyzed		ring ration	of a	dition cid mple		ring stion
No.‡	Description‡	A #	B #	A #	В #	A #	В#
21	Greenhouse soil (water extract)		_	_			
22	Mushroom compost (cold-water extract)		_	_		Barrella	_
23	Mushroom compost (autoclave extraction)	_			_		_
24	Mushroom compost extract plus KNO <sub>3</sub> solution	<b>I</b>	_	_	_		
25	Aspergillus niger, residual soln plus KNO <sub>3</sub> soln.	_		_			_
26	Fusarium culmorum, residual soln. plus KNO <sub>3</sub> soln.	_	_	_		_	_
27	Crude peat extract (boiled)	_	_	_	_	_	_
28	Sphagnum moss (H <sub>2</sub> O extract)						
30	Selaginella, expressed sap						_
31	Selaginella (alcohol extract)		_			_	-
32	Selaginella (water extract)			_		_	
33	Barley plants (green) plus 10 cc. residual soln. (high in nitrate- N)	_	_				
34	Same as No. 33 except much lower in nitrate-N				4-		_
35	Wheat plants (green) plus 10 cc. residual soln. (high in nitrate-N)		_				

### TABLE II—Continued

			Quali	tative 1	tests ma	det II	
Sample analyzed		during evaporation		on addition of acid to sample		during digestion	
No.‡	Description‡	A #	B #	A #	B #	A #	B #
36	Same as No. 35 except much lower in nitrate-N	_					
37	Pea plants (green) plus 10 cc. residual solution. (high in nitrate-N)						
38	Same as No. 37 except much lower in nitrate-N		_	_		_	
39	Residual soln. from wheat cultures containing 200 mgs. nitrate-N						
40	Same as No. 39 except containing 100 mgs. nitrate-N		_	_	_		_
41	Same as No. 39 except containing 50 mgs. nitrate-N			_	_		
42	Same as No. 39 except containing 25 mgs. nitrate-N				_	_	
43	Same as No. 39 except containing 10 mgs. nitrate-N		_		_	_	
44	Same as No. 39 except containing 5 mgs. nitrate-N		_		_		_
45 46 47 48 49 50	These samples were the same as those of Nos. 39–44 except that barley plants were used instead of wheat						
52	Alanin plus KNO <sub>3</sub> solution					_	_
53	Asparagin plus KNO <sub>2</sub> soln.					_	_
54	Sugar-cane extract plus KNO <sub>3</sub> solution			_	_	_	_
60	Sugar-beet (water extract)	_			_	_	_

<sup>\*</sup>Ranker ('25).
† The methods of conducting these tests are given under "Methods."
|| The analysis of each separate sample was repeated 5 times; all 5 tests were in agreement, and as indicated for each sample, respectively.
‡ The sample numbers correspond to those numbers used in the discussion of

materials.

<sup># &</sup>quot;A" is used to designate the diphenylamine test; "B," the test with Nessler's ceagent.

A critical examination of certain methods for the determination of nitrate-nitrogen has been undertaken in this investigation. This phase of the subject is still under investigation. It seems desirable, however, to include in this report certain results which were obtained. The accuracy of the Kjeldahl-Gunning-Arnold method (Association of Official Agricultural Chemists, '25, page

TABLE III

ACCURACY OF THE KJELDAHL-GUNNING-ARNOLD METHOD\* FOR THE DETERMINATION OF "TOTAL NITROGEN NOT INCLUDING NITRATE NITROGEN"

(Expressed as cc. N/50 HCl neutralized by the ammonia content of the distillate)

Sample determined		Total N		"Total ni not incl nitrate		
No.†		present including nitrate-N	Nitrate-N   present	Amt. present	Amt. found	% error‡
52	Alanin, KNO <sub>1</sub> soln. sample in solution. sample in solution. sample dry, evaporated.	$29.3 \pm .04$	17.1 ± .01	$12.2 \pm .04$	13.5 13.0 12.7	+ 10.7 + 6.6 + 4.1
53	Asparagin, KNO <sub>1</sub> soln. sample in solution sample in solution.	$27.7 \pm .06$	17.1 ± .01	10.6 ± .06	11.9 12.2	+ 12.3 + 15.1
24	Mushroom compost, KNO <sub>3</sub> . sample in solution. sample in solution. sample dry, evaporated.	25.4 ± .06	17.1 ± .01	8.3 ± .06	9.0 8.7 10.8	+ 8.4 + 4.8 + 30.1
54	Sugar cane, KNO <sub>3</sub> soln. sample in solution. sample in solution. sample dry, evaporated.	$49.5 \pm .05$	17.1 ± .01	32.4 ± .05	38.5 36.8 41.9	+ 18.8 + 13.6 + 29.3
25	Aspergillus niger, KNO <sub>3</sub> . sample in solution. sample in solution. sample dry, evaporated.	17.8 ± .04	17.1 ± .01	0.7 ± .04	0.9 1.1 2.3	+ 28.6 + 57.1 + 228.6
26	Fusarium culmorum, KNO <sub>3</sub> . sample in solution. sample in solution. sample dry, evaporated.	18.3 ± .05	17.1 ± .01	1.2 ± .05	1.3 2.1 3.2	+ 8.3 + 75.0 + 166.7

<sup>\*</sup> Association of Official Agricultural Chemists ('25), page 8, No 24.

<sup>†</sup> The sample numbers correspond to those numbers used in the discussion of materials.

<sup>||</sup> This amount of nitrate-nitrogen was added as 25 cc. of solution No. 74; the other component of the sample was nitrate-free as indicated by a negative diphenylamine test.

<sup>†</sup> The basis of these calculations was the amount of nitrogen present (column No. 3) in the form of "total nitrogen not including nitrate-nitrogen."

8, No. 24) for the determination of "total nitrogen not including nitrate-nitrogen" is well established. In the analysis of plant materials (in which nitrate-nitrogen is usually present) the accuracy of this method has been assumed rather generally. Its applicability to samples of plant materials has not always been satisfactorily demonstrated. In attempting to investigate the possibility of estimating nitrate-nitrogen, by finding the difference between the modified official method and the Kjeldahl-Gunning-Arnold method, it seemed imperative to determine the accuracy of the latter method when applied to plant materials. The data of table III cover a part of this work.

The outstanding facts obtained from the data of table III are that all the results are high, and that in the presence of nitratenitrogen this method is not accurate when applied to plant materials. Other data, not included in this report, support this conclusion and support the suggestion made by Strowd ('20) that "appreciable amounts of nitrate were apparently reduced without zinc and salicylic acid." In all cases the plant extract component of the respective samples was known to be definitely free from nitrates and nitrites, so that the 25 cc. of solution No. 74 (KNO<sub>3</sub>) furnished the only nitrate-nitrogen present. In the case of samples Nos. 25 and 26, the experimental error is high due to the low content of organic nitrogen, and the calculation of the "% error" therefore is subject to the same magnitude of error. As a result these data (Nos. 25 and 26) are not entirely satisfactory. It is of interest to note that in all cases, except for sample No. 52, the inaccuracy of the method is greatly increased when the sample is dry. This result is in sharp contrast to the condition of the official salicylic-thiosulphate method (Ranker, '25) in which case low results were obtained unless the sample was dry.

The first attempts to determine nitrate-nitrogen only, by finding the difference between the modified official method and the Kjeldahl-Gunning-Arnold method, were made on samples similar to those included in table III. Later determinations confirmed the earlier results, but only the latter are included in table IV.

The data of table IV support the same conclusions as do those of table III. This condition would be expected from the situa-

TABLE IV DETERMINATION OF NITRATE-NITROGEN BY FINDING THE DIFFERENCE BETWEEN THE MODIFIED OFFICIAL METHOD AND THE KJEL-DAHL-GUNNING-ARNOLD METHOD\*

No.	Sample analyzed  Description	Nitrate-N present in sample (mgs.)	Total N found by modified official method (mgs.)	N found by K-G-A. method (mgs.)	Nitrate-N found by difference (mgs.) [compare with column I]	% error**
65	Sugar-beet, KNO <sub>3</sub>	4.8 ± .01	15.5 ± .03	11.3 11.6 11.8	4.2 3.9 3.7	12.5† 18.8† 22.8‡
54	Sugar-cane, KNO <sub>3</sub>	4.8 ± .01	13.9 ± .01	10.8 10.3 11.7	3.1 3.6 2.2	35.4† 24.9† 54.2‡
24	Mushroom compost plus KNO <sub>3</sub>	4.8 ± .01	7.1 ± .02	2.5 2.4 3.0	4.6 4.7 4.1	- 4.2† - 2.1† - 14.6‡
25	Aspergillus niger plus KNO <sub>3</sub>	4.8 ± .01	5.0 ± .01	0.3 0.3 0.6	4.7 4.7 4.4	- 2.1† - 2.1† - 8.3‡
26	Fusarium culmorum plus KNO <sub>2</sub>	4.8 ± .01	5.1 ± .01	0.4 0.6 0.9	4.7 4.5 4.2	- 2.1† - 6.2† - 12.5‡
52	Alanin, KNO₃	4.8 ± .01	8.2 ± .01	3.8 3.6 3.6	4.4 4.6 4.6	- 8.3† - 4.2† - 4.2‡
53	Asparagin, KNO,	4.8 ± .01	7.8 ± .02	3.3 3.4	4.5 4.4	- 6.2† - 8.3‡

\*\* The basis of this calculation was the amount of nitrate-nitrogen present in the sample (column I), that is,  $4.8 \pm .01$  mgm. nitrate-nitrogen.

tion as noted in the latter case. There seems to be some correlation between the inaccuracy of the method and the presence of certain reducing substances. Certain results which have been obtained, but which are not included in this report, seem to warrant the belief that with certain modifications these difficulties can be overcome and accurate results obtained in the presence of nitrate-nitrogen. With this accomplished it seems very probable that nitrate-nitrogen only can be quantitatively

<sup>\*</sup> Association of Official Agricultural Chemists ('25), page 8, No. 24. || This amount of nitrate-nitrogen was added as 25 cc. of solution No. 74; the other component of the sample was nitrate-free as indicated by a negative diphenyl-

<sup>†</sup> Sample in solution, that is, just as made up. ‡ Sample first evaporated to dryness on a water bath under vacuum.

determined by finding the difference between the modified official method and a modification of the Kjeldahl-Gunning-Arnold method.

Early in this investigation it became apparent that certain details of manipulation influenced, more or less, the accuracy of the results obtained. In some cases the procedure followed was determined by these factors. The methods of qualitative controls which were developed (discussed under "Methods") made it possible to determine, accurately, the influence of certain details of procedure. Many of the samples were subjected to extreme conditions in order to determine the limits to which certain procedures could be carried. The data obtained from these various studies are given in tables v-Ix.

Certain points in regard to the data of table v should be mentioned. The neutrality of the sample is a very important consideration with most of the samples, Nos. 5 and 5b, 29 and 32, 17 and 21. For such samples, this one factor of neutrality would determine the accuracy of subsequent quantitative The samples just referred to were on the acid side of neutrality prior to neutralization. On the other hand, when the reaction is alkaline a loss of ammonia may occur (sample No. 26a). Another effect of this factor of neutrality should be noted as evidenced by samples Nos. 5b and 5c, 32 and 32a. Sample No. 5b, Phoma Betae, having a PH value of 6.5, could be quantitatively evaporated just to dryness when neutralized, but when evaporated to ash dryness there was a loss of nitrogen (sample No. 5c). On the other hand, sample No. 32a, having a Ph value of 6.8, could be evaporated to ash dryness without a loss of nitrogen. Of all the materials analyzed throughout this entire investigation Phoma Betae was the most difficult. When the factors involved (adjustment to neutrality and evaporation just to dryness) were controlled, however, no difficulty was experienced as is shown by the quantitative results obtained (sample No. 5, table 1). Sample No. 25 illustrates the opposite extreme; this sample (Aspergillus niger) having a P<sub>H</sub> value of 3.9 required no adjustment to neutrality even in the presence of added nitrate-nitrogen. These two materials (Nos. 5 and 25) represent the extremes met with. Somewhat similar variations were observed throughout the entire

TABLE V INFLUENCE OF NEUTRALITY OF THE SAMPLE UPON THE STABILITY OF NITROGEN DURING THE PROCESS OF EVAPORATION

		Qual. tests for loss of N durin evaporation		
No.*	Sample analyzed and method of treatment	A #	В #	
5	Phoma Betae residual soln., high in NO <sub>3</sub> , P <sub>B</sub> 6.5 (not adjusted to neutrality, evaporated just to dryness)	+		
5a	Same as No. 5, except evaporation was less vigorous	+		
<b>5</b> b	Same as No. 5, except sample adjusted to neutrality		_	
<b>5</b> c	Same as No. 5b, except sample evaporated to ash dryness	+		
29	Selaginella extract, 14 ds. old. P <sub>H</sub> 5.5, not neutralized (evaporated just to dryness)	+		
32	Selaginella extract, used immediately, P <sub>H</sub> 6.8 (evaporated just to dryness)		_	
32a	Same as No. 32, except sample evaporated to ash dryness		_	
26	Fusarium culmorum residual soln., plus KNO <sub>3</sub> , P <sub>H</sub> 8.5	_		
26a	Same as No. 26, except trace NH <sub>3</sub> added, P <sub>H</sub> 8.7		±	
17	Greenhouse soil with mushroom compost, H <sub>2</sub> () extract, stood in lab. 10 ds., P <sub>H</sub> 5.7 (evap. just to dryness)	+		
17a	Same as No. 17, except extract autoclaved before testing	+	_	
21	Sample made from same material as was No. 17, extract analyzed immediately, $P_{\rm H}$ 7.1	_	_	
25	Aspergillus niger residual soln. plus KNO <sub>3</sub> , P <sub>H</sub> 3.9 (adjusted to neutrality and evap. just to dryness)	_		
25a	Same as No. 25, except evaporated to ash dryness	- 1		
25b	Same as No. 25, except sample not neutralized (P <sub>H</sub> 3.9)	-	_	
25c	Same as No. 25a, except sample not neutralized (P <sub>H</sub> 3.9)	) <u> </u>		

<sup>\*</sup> The sample numbers correspond to those numbers used in the discussion of materials.

list of materials used; some required adjustment to neutrality and others did not.

<sup>†</sup> The methods of conducting these tests are given under methods.

# "A" is used to designate the diphenylamine test; "B," to designate the test with Nessler's reagent.

<sup>‡</sup> The analysis of each separate sample was repeated 5 times; all 5 tests were identical and as indicated for each sample, respectively.

The method of adjusting a sample to neutrality was found to be of extreme importance to accurate procedure. The influence of this factor is apparent from a consideration of the data of table vi.

TABLE VI INFLUENCE OF METHOD OF NEUTRALIZING THE SAMPLE UPON THE STABILITY OF NITROGEN DURING THE PROCESS OF EVAPORATION

		Nitrogen found in sample	Qual. testst for loss of N during evaporation	
	Sample analyzed and method of treatment*	(mgs.)	A #	В#
(a)	Shive's nutrient soln., calculated to contain 200 mgs. nitrogen per 950 cc. (indicator; added to sample, adjusted to neutrality directly)		+	_
(b)	Same as (a), except indicator was omitted; adjusted to neutrality by adding a predetermined amount of alkali	202.7		
(c)	Shive's nutrient soln., calculated to contain 100 mgs. nitrogen per 950 cc. (indicator added to sample, adjusted to neutrality directly)	96.3	+	
(d)	Same as (c), except indicator was omitted; adjusted to neutrality by adding a predetermined amount of alkali	101.3		
(e)	Shive's nutrient soln., calculated to contain 50 mgs. nitrogen per 950 cc. (indicator added to sample, adjusted to neutrality directly)	47.9	+	
(f)	Same as (e), except indicator was omitted; adjusted to neutrality by adding a predetermined amount of alkali	48.4		

The data of table vi need little comment; the same qualitative results were obtained with other samples and other indicators. Phoma Betae samples were extremely difficult to deal with. choice of indicators that may be used is limited to those which contain no nitrogen, when the indicator is added directly to the sample.

In the procedure for the modified official method (Ranker, '25, p. 371) it is recommended that the sample be neutralized

<sup>\*</sup> All samples were evaporated just to dryness.
† The methods of conducting these tests are given under "Methods."
\* "A" is used to designate the diphenylamine test; "B," the test with Nessler's

The indicator used for these data was brom-cresol purple.

and then evaporated under vacuum just to dryness. question of neutrality has been considered in connection with the data of tables v and vi. The study of the factors involved in the process of evaporation was continued, to ascertain the limits of the process and to evaluate the recommendation that the sample be evaporated just to dryness, when applied to various samples. A portion of the results obtained are reported in table vii; the book of data, from which these were taken, contains many more just like those included in table vii. The data included are entirely representative and a sufficient number were taken to show the extremes and the range of variation in the results obtained. In all such analyses the factor of neutrality of the sample was taken care of and controlled; if the sample required neutralization (sample 5, for example) it was adjusted to neutrality by adding a predetermined amount of acid or alkali; if the sample did not require neutralization (samples 33 and 9, for example) it was not neutralized. The data obtained, therefore, may be compared directly in relation to the process of evaporation under vacuum. All of the samples used contained nitratenitrogen, either present as shown by a positive diphenylamine test, or as added nitrate-nitrogen, in which case 25 cc. of solution No. 74 was added per sample.

The outstanding fact illustrated by the data of table vii is that the process of evaporation cannot be conducted in any "haphazard" manner. Some samples require rather careful evaporation just to dryness while others may be evaporated to ash dryness and heated for an hour afterward without loss of nitrogen. The most stable material used was No. 3 (Aspergillus niger), which with added nitrate-nitrogen (sample No. 25a) could be evaporated to ash dryness with safety. In all cases when the sample was evaporated to partial dryness only, there was a subsequent loss of nitrogen at the time the salicylic acid mixture was added and also during the process of digestion (illustrated by samples No. 33, 35, 37, etc.). The influence of the presence of water in the sample is demonstrated, further, by a consideration of the data for samples Nos. 17 and 17a; these samples were evaporated just to dryness and the acid mixture was added: 5 minutes later a small amount of water was added

## TABLE VII

INFLUENCE OF THE EXTENT TO WHICH EVAPORATION IS CARRIED UPON THE STABILITY OF NITROGEN DURING THE PROCESSES OF EVAPORATION, ADDITION OF THE ACID MIXTURE, AND SUBSEQUENT DIGESTION

		Qual. tests for loss of nitrogen†					†
Sample analyzed*		during evaporation		on addition of acid to sample		during digestion	
No ‡	Description‡	A #	B #	A #	B #	A #	B #
33	Barley plants plus 10 cc. residual soln. (evap. to partial dryness)		_	+		+	
33a	Same as No. 33, except evaporated just to dryness			_			_
33b	Same as No. 33, except evaporated to ash dryness	+		_			
35	Wheat plants plus 10 cc. residual soln. (evap. to partial dryness)			+	_	+	_
35a	Same as No. 35, except evaporated just to dryness		_				
<b>35</b> b	Same as No. 35, except evaporated to ash dryness	+					
37	Pea plants plus 10 cc. residual soln. (evap. to partial dryness)			+		+	
37a	Same as No. 37, except evaporated just to dryness						
<b>37</b> b	Same as No. 37, except evaporated to ash dryness	+			_		_
17	Greenhouse soil plus mushroom compost, H <sub>2</sub> O extract (evap. just to dryness; acid mixture added; 5 minutes later H <sub>2</sub> O added)	_		+		+	
17a	Same as No. 17, except H <sub>2</sub> O added 24 hrs. after acid mixture			+	_	+	
17b	Same as No. 17, except evap. to ash dryness, no water added	+					
20	Garden pea, water extract (evap. just to dryness)	_			_		
20a	Same as No. 20, except evaporated to ash dryness	+					_
5	Phoma Betae residual soln. (evap. just to dryness)	<u> </u>			-0		

#### TABLE VII—Continued

		Qual. tests for loss of nitrogen†				†	
Sample analyzed*		during evaporation		on addition of acid to sample		during digestion	
No.‡	Description‡	A #	В #	A #	В#	A #	В #
5a	Same as No. 5, except evaporated to ash dryness	+	_		_	_	_
<b>5</b> b	Same as No. 5, except evap. to partial dryness			+		+	
26	Fusarium culmorum residual soln. plus KNO <sub>3</sub> , evap. just to dry- ness			_			
26a	Same as No. 26, except evaporated to ash dryness	_		_	_		
26b	Same as No. 26, except evap. to partial dryness	_	_	+	_	+	_
25	Aspergillus niger residual soln., plus KNO <sub>3</sub> , evap. just to dry- ness			_		_	_
25a	Same as No. 25, except evap. to ash dryness	_			_	_	
25b	Same as No. 25, except evap. to partial dryness			+		+	
9	Celery, water extract, evap. just to dryness		_				
9a	Same as No. 9, except evap. to ash dryness		_	_	_	_	_
24	Mushroom compost extract plus KNO <sub>3</sub> , evap. just to dryness	- 1			_	_	_
24a	Same as No. 24, except evap. to ash dryness				_	_	

<sup>\*</sup>The analysis of each separate sample was repeated 5 times; all 5 tests were identical and as indicated for each sample, respectively.

† The methods of conducting these tests are given under "Methods."

and there was a loss of nitrogen; 24 hours later a small amount of water was added to duplicate samples and there was a similar loss of nitrogen. Special attention is called to sample No. 9

The sample numbers correspond to those numbers used in the discussion of materials.

<sup># &</sup>quot;A" is used to designate the diphenylamine test; "B," the test with Nessler's reagent.

(celery). This material was very high in nitrate-nitrogen, second only to sample No. 32 (Selaginella apus). Both of these samples required no neutralization and could be evaporated to ash dryness without a loss of nitrogen (since the results were identical the data for sample No. 9 only are reported in this connection).

A special phase of the process of evaporation is illustrated by such materials as whole green plants. When the sample involved contains whole green plants, plus about 10 cc. of the nitrate-containing residual solution, considerable difficulty is experienced in evaporation under vacuum: (1) the colloidal complex of the plant does not give up its water content rapidly in the humid atmosphere inside the flask; (2) the small amount of moisture present in the sample does not evolve sufficient steam to break down effectively the plant structure; (3) the steam evolved under such conditions does not heat the flask sufficiently to prevent condensation and consequent run-back. The semiequilibrium obtained, however, is such that evaporation may be accomplished in this manner but the time required is too long for efficiency. It required 3-6 hours to evaporate the sample to partial dryness under vacuum, 6-8 hours to evaporate it just to dryness under vacuum, and 7-10 hours, to full dryness under vacuum. These time intervals refer only to the particular kind of sample mentioned above, namely, whole green plants plus about 10 cc. of the nitrate-containing residual solution. Furthermore, it is difficult to judge, under these conditions, when the sample is evaporated just to dryness (sample No. 35, table VIII); if carried on just past this stage there is a loss of nitrogen (sample No. 35a), especially if the sample is high in nitrate-nitrogen. These difficulties may be overcome in one of two ways: (1) by the addition of an appreciable amount (50-100 cc.) of distilled water, sufficient to evolve steam enough to break down completely the plant structure and organization; (2) by the use of some such ventilation device as that illustrated by fig. 3, by which a continuous stream of dry air is used to carry off the vapors from the sample. This latter method is recommended. With this device there was no loss of nitrogen from any sample of whole green plants, even when large amounts of nitrate-nitrogen were added. Some of the data obtained from

this study are reported in table viii, and are representative and typical of all the data obtained.

Certain points in connection with the data of table VIII are worthy of mention. The time periods required to evaporate the samples under vacuum have been given, namely: to evaporate the samples, under vacuum, to partial dryness, 3-6 hours; just

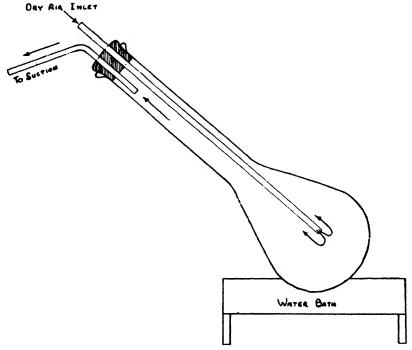


Fig. 3. Arrangement of apparatus for the evaporation of whole green plants by dry-air ventilation.

to dryness, 6-8 hours; to full dryness, 7-10 hours. In contrast, the time periods required to evaporate similar samples to comparable stages of dryness, by using the special ventilation device (fig. 3), were 10-15 minutes, 12-20 minutes, and 15-25 minutes, respectively. When this latter method was used to evaporate such samples there was no loss of nitrogen in any of the tests made. Sample No. 35c, for example, was heated for 1 hour after it was completely dry, with no loss of nitrogen.

Very early in this investigation it was noticed that certain irregular results in titration were obtained. For example, in the

#### TABLE VIII

INFLUENCE OF THE METHOD OF DRYING WHOLE GREEN PLANTS UPON THE STABILITY OF NITROGEN DURING THE PROCESSES OF EVAPORATION, ADDITION OF THE ACID MIXTURE, AND SUBSEQUENT DIGESTION

		1	==					<del></del>				
		Qual. tests for loss of nitrogen†										
Sample analyzed				ing ation‡	of a	dition scid mple†	during digestion‡					
No.*	Description*	A #	!	В #	A #	В #	A #	В#				
35	Wheat plants plus 10 cc. of residual solution high in nitrate- nitrogen evaporated under vacu- um just to dryness			_			_	_				
35a	Same as No. 35 except evaporated under vacuum to full dryness	+					_					
35b	Same as No. 35 except evaporated under vacuum to partial dryness				+		+					
35c	Same as No. 35 except evaporated just to dryness by a special ventilation device**	_										
35d	Same as No. 35c except evaporated to full dryness	_			[	_						
35e	Same as No. 35c except heated for 1 hour after sample was completely dry				_							

<sup>\*</sup>The sample numbers correspond to those numbers used in the discussion of materials.

\*"A" is used to designate the diphenylamine test; "B," the test with Nessler's

\*\* See fig. 3.

simple determination of nitrogen present in a known solution of ammonium sulphate, 2 each of the 6 titrations consistently required more alkali to titrate the excess acid to neutrality than did each of the other 4; also, these same 2 flasks contained a more or less milky distillate that was quite distinctive from the crystal-clear distillate of the others. After several attempts to locate the cause of this difficulty it was noticed that whenever a certain grade of rubber tubing was used, to connect the condenser bulb with the block tin condenser tube of the distillation apparatus,

aterials.
† The methods of conducting these tests are given under "Methods."

<sup>†</sup> The same results were obtained for barley and pea plants, samples No. 33, 34, 37, 38, and for wheat sample No. 36. Each test was repeated 5 times for each separate sample and the same results were obtained each time.

TABLE IX INFLUENCE OF RUBBER TUBING CONNECTIONS\* UPON THE QUANTITATIVE DISTILLATION OF AMMONIA INTO STANDARD ACID

No.t	Sample analyzed and nature of rubber tubing connections used  Description	Cc. N/50 acid neutralized by ammonia contained in distillate	Error (cc. N/50 acid)
NO.	Description	III distinate	aciu)
11	Tomato fruits, water extract Distillation through connection No. 2; average of 4 tests   Distillation through connection No. 1‡ used on condenser bulb No. 3	$39.1 \pm .08$ $37.6$	1.5
9	Celery, water extract Distillation through connection No. 2; average of 3 tests   Distillation through connection No. 1; used on condenser bulb No. 3 used on condenser bulb No. 4	51.9 ± .04 50.9 50.0	— 1.0 — 1.9
17	Greenhouse soil and mushroom compost, H <sub>2</sub> O extract Distillation through connection No. 2; average of 4 tests   Distillation through connection No. 3 # used on condenser bulb No. 5		4.5
73	Heavy clay loam soil, water extract Distillation through connection No. 2, average of 4 tests   Distillation through connection No. 1‡ used on condenser bulb No. 1 used on condenser bulb No. 2 used on condenser bulb No. 3 used on condenser bulb No. 4	3.2 ± .03  2.6 2.9 2.9 2.6	0.6 0.3 0.3 0.6
18	Sugar-cane, water extract Distillation through connection No. 2; average of 5 tests   Distillation through connection No. 1‡ used on condenser bulb No. 1 used on condenser bulb No. 2 used on condenser bulb No. 3 used on condenser bulb No. 4 used on condenser bulb No. 5	44.7 ± .06 43.9 43.1 42.9 40.9 44.4	0.8 1.6 1.8 3.8 0.3

<sup>\*</sup> As used in this table, the term "connection" refers only to that length of rubber tubing used to connect a glass condenser bulb to a block tin condenser tube of the Kjeldahl distillation apparatus.

† The sample numbers correspond to those numbers used in the "Discussion of materials.

cm. of the total length was exposed to ammonia vapors during distillation.

Connections No. 1 were such that about 6 cm. of the total length was exposed to ammonia vapors during distillation. Before use the rubber was extracted once with N/10 NaOH, washed in dilute HCl, and rinsed thoroughly. The rubber tubing used was the ordinary white laboratory grade, thick-walled, cloth-wrapped (A. H. Thomas' No. 8830).

[Connections No. 2 were such that not more than 0.5 cm. of the total length was

exposed to ammonia vapors during distillation. Before use the rubber was treated as in the case of connections No. 1. The rubber tubing used was A. T. Thomas' No. 8834: black gum and sulphur; steam-cured; thick-walled; smooth bore; of fairly good quality. These connections were adopted for all subsequent work.

# Connections No. 3 were the same as connections No. 1 except that about 20

these milky distillates were obtained. In such distillates the calculated nitrogen content was invariably low. The particular grade of rubber tubing which gave these results was that described in the footnote to table IX and was used in making the "connections No. 1" as indicated. When extracted with N/10 sodium hydroxide a yellow-green sodium sulphide mixture was obtained even after 6 such extractions. This grade of rubber is entirely unsatisfactory for such connections. A better grade of rubber tubing was obtained; this is described in the footnote to table IX and was used to make "connections No. 2" as indicated. (Note that the total length of rubber exposed to the ammonia fumes was not more than .5 cm.). The first extraction of this rubber with N/10 sodium hydroxide produced the characteristic yellow-green sodium sulphide mixture but all subsequent extractions were free from this material. Six check distillations of ammonia, through connections No. 2, produced a probable error of +.0045; these results were entirely satisfactory and this grade of rubber tubing was adopted for all subsequent work. Certain few analyses were made to illustrate the influence of these different rubber connections and these data are given in table ix.

### GENERAL DISCUSSION AND CONCLUSIONS

There are certain aspects of the data presented which require some discussion and which require correlation with the data previously reported (Ranker, '25) and with the larger aspects of the determination of nitrogen. In the determination of any form of nitrogen it would seem logical, first, to obtain a method which is accurate for the determination of total nitrogen. Two methods are used generally: (1) some modification of the Devarda method, and (2) the official salicylic-thiosulphate method (Association of Official Agricultural Chemists, '25, p. 9, No. 29). The first method is time-consuming; there is a preliminary distillation with Devarda's alloy, an acid digestion, and a second alkaline distillation. The second method is not accurate when applied to most plant materials in the presence of water (Ranker, '25). This method, however, is relatively rapid when compared with the Devarda method. It is considered that the inaccuracies

of this second method (official salicylic-thiosulphate method) have been overcome, with no loss of rapidity, by the procedure for the modified official method (Ranker, '25). The data reported in this paper are offered in further support of the accuracy of this method.

Certain aspects of the chemistry of the modified official method should be considered at this point. Roughly, the nitrogen content of all materials may be divided into 2 classes: (1) organic, and (2) inorganic. Either of these may have an acid or basic reaction. The data of this investigation show that the acid or basic nature of the various forms of nitrogen, as such, does not seem to influence the stability of the total nitrogen contained in the sample. On the other hand, the reaction of the sample seems to determine the stability of its nitrogen-content in many cases. Both acid and basic forms of nitrogen may be present in rather equal proportions in the same sample. Such an occurrence may explain why it is necessary to adjust certain samples to neutrality prior to evaporation Such an adjustment to neutrality may exert a stabilizing influence, due directly to the relative decrease of hydrogen- or hydroxyl-ions. It may exert, also, an indirect influence, due to the formation of new nitrogen compounds which are more stable than those forms of nitrogen present prior to adjustment of the sample to neutrality.

In regard to the organic nitrogen which is present, it would be transformed into ammonia-nitrogen very much as would occur with the Kjeldahl-Gunning-Arnold method and need be considered no further here. The inorganic nitrogen content of a sample, however, presents a different situation. Of the various forms of inorganic nitrogen, nitrate-nitrogen commands most attention. Given the proper condition of dryness (evaporation just to dryness) for a nitrate-containing sample, the reduction of nitrate-nitrogen to ammonia-nitrogen may be considered to take place somewhat as follows, with the use of salicylic acid:

# (1) $NaNO_3 + H_2SO_4 \rightarrow NaHSO_4 + HNO_3$

OH COOH 
$$+ 0$$
 N  $- OH$   $+ 0$  NO<sub>1</sub>

OH COOH  $+ 6H$   $+ 6H$  Sodium thiosulphate action  $+ 2H_1O$ 

OH COOH

NO<sub>1</sub>

OH COOH

 $+ 6H$  Sodium thiosulphate action  $+ 2H_1O$ 
 $+ 2H_1O$ 

OH COOH

 $+ 6H$   $+ 100$ 
 $+ 2H_1O$ 
 $+ 2H_$ 

By equation (2) it is evident that nitric acid is responsible for the nitrification of the salicylic-acid molecule. The nitric acid is obtained from the nitrates present in the sample (equation 1) by the action of the sulphuric acid which is present in the salicylic-acid mixture (1.0 gm. salicylic acid to 30 cc. sulphuric acid). The sulphuric acid plays another important rôle. The quantitative completion of equation (2) and equation (3) toward the right depends upon the loss of water. For each atom of nitrogen thus combined 3 molecules of water are formed. This water is removed from the reaction by the desiccating action of the sulphuric acid. The reactions (equations 2 and 3), thereby

continue to quantitative completion and there is no loss of nitrogen, providing the sample was first evaporated to dryness (Ranker, '25). The sulphuric acid functions in yet another manner, namely, in the digestion of the amino-salicylic-acid molecule and the formation of ammonium sulphate (equation 4).

The various steps in the procedure recommended for the modified official method have been reinvestigated and the results obtained do not warrant any alterations in the procedure. Certain qualifying conditions must be recognized, however: (1) some samples must be adjusted to neutrality prior to evaporation while other samples require no such adjustment; (2) some samples must be carefully evaporated just to dryness and other samples may be roughly evaporated to ash dryness without a loss of nitrogen (the particular chemical or physical complexes responsible for these differences are not known); (3) in the distillation process paraffin may be omitted in some cases but must be present in others to prevent foaming. In just which cases it is safe to take advantage of these qualifying conditions, in order to simplify any step in the procedure, must be decided by the individual investigator using the method. Inasmuch as these factors (neutralization and evaporation just to dryness) are essential to the accurate determination of total nitrogen in some samples they cannot be omitted from the general procedure recommended (Ranker, '25, p. 371).

The above considerations lead directly to a consideration of the importance of the qualitative tests. Without some such tests it is impossible to control, accurately, the various factors involved in the quantitative determination of nitrogen by any of the acid digestion methods, impossible to know, definitely, that there has or has not been a loss of nitrogen in the process, and very difficult to locate any source of error that might exist. It is strongly recommended that suitable qualitative tests (those used in this investigation have been found satisfactory) be considered as an integral part of nitrogen-determination methods whenever it is at all feasible to use them. An example or two might be of value to illustrate this suggestion: (1) certain of the factors contributing to the inaccuracies of methods investigated by Mitscherlich and Herz ('09) could have been definitely located

and corrected in this manner; (2) had such tests been used Strowd ('20), using samples containing plant materials plus nitrate-nitrogen in solution, probably would have detected the inaccuracy of the "Kjeldahl method modified to include nitrate" under such conditions; (3) the use of such qualitative tests would have deterred Gallagher ('23, p. 67) from his theoretical denunciation of the principle of reduction of nitrates in acid medium; (4) the data presented in this report are filled with instances in which all sorts of results would have been obtained had not the quantitative data been checked with qualitative tests on the procedure used. Other examples of a similar nature are abundant. It is fortunate, indeed, that the various methods for the determination of nitrogen are so well adapted to the use of qualitative control tests for the loss of nitrogen. It is unfortunate that these qualitative tests are so seldom used.

One other aspect of this investigation should be mentioned at this time, namely, the determination of nitrate-nitrogen. Consideration of this subject will be restricted to the determination of nitrate-nitrogen by finding the difference between the Kieldahl-Gunning-Arnold method and the modified official method (a separate report is being prepared on certain phases of the determination of nitrate-nitrogen in plants by the Devarda method). The inaccuracies of the official salicylic-thiosulphate method (Association of Official Agricultural Chemists, '25, p. 9, No. 29) in the presence of moisture have prevented the determination of nitrate-nitrogen by finding the difference between that method and the Kieldahl-Gunning-Arnold method. With the more accurate procedure of the modified official method such a determination of nitrate-nitrogen was thought possible. The inaccuracies of the Kjeldahl-Gunning-Arnold method when applied to samples containing nitrates (data of table III), however, prevent this possibility. The data presented in this report include only negative results in this direction. Certain tentative modifications which have been tried, however, indicate the possibility of perfecting the Kjeldahl-Gunning-Arnold method to the point that "total nitrogen not including nitrate-nitrogen" only, can be quantitatively determined in the presence of nitratenitrogen. With this accomplished it is very probable that

nitrate-nitrogen only can be quantitatively determined by finding the difference between the modified official method and a certain modification of the Kjeldahl-Gunning-Arnold method.

The various data which have been obtained in this investigation seem to warrant the following conclusions:

- 1. The modified official method (Ranker, '25) is accurate for the determination of total nitrogen in soil extracts and in samples of biological materials, on the plant side.
- 2. The Kjeldahl-Gunning-Arnold method (Association of Official Agricultural Chemists, '25, p. 8, No. 24) for the determination of "total nitrogen not including nitrate nitrogen" is not accurate in the presence of nitrate-nitrogen. Until this method is perfected, so that it is accurate in the presence of nitrate-nitrogen, it is useless to attempt the determination of nitrate-nitrogen only, by finding the difference between the modified official method and the present Kjeldahl-Gunning-Arnold method.
- 3. The data obtained do not warrant any alteration in the recommended procedure for the modified official method (Ranker, '25). The stability of the nitrogen complex of certain samples may permit of certain deviations from the recommended procedure, however. This is a matter for individual judgment and will be governed by the particular samples under investigation. These deviations can be made safely, only when suitable qualitative tests are used to detect a loss of nitrogen from the sample.
- 4. Suitable, accurate qualitative tests for the loss of nitrogen should be as much an integral part of the various quantitative nitrogen methods as the actual quantitative determination itself, whenever it is possible to use them without decreasing the accuracy of the quantitative method involved.

## SUMMARY

- 1. The accuracy of the modified official method for the determination of total nitrogen, as previously reported, is verified.
- 2. The application of the modified official method is extended to a wide variety of samples, including soil extracts and samples of biological materials, on the plant side.
- 3. It is demonstrated that the Kjeldahl-Gunning-Arnold method, for the determination of "total nitrogen not including

nitrate-nitrogen," is not accurate in the presence of nitrate-nitrogen.

- 4. Until the Kjeldahl-Gunning-Arnold method is modified to be accurate in the presence of nitrate-nitrogen, it is useless to attempt the determination of nitrate-nitrogen by finding the difference between this method and the modified official method.
- 5. Certain details of manipulation and procedure are discussed, and their influence on the accuracy of methods for nitrogen determination is demonstrated.
- 6. The value of suitable qualitative tests, for loss of nitrogen, to be used as an integral part of quantitative methods for the determination of nitrogen, is demonstrated, discussed, and the use of such tests is recommended.

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# SOME CYTOLOGICAL AND PHYSIOLOGICAL STUDIES OF MOSAIC DISEASES AND LEAF VARIEGATIONS<sup>1</sup>

#### FANNY FERN SMITH

Instructor in Botany, Lindenwood College Formerly Rufus J. Lackland Research Fellow in the Henry Shaw School of Botany of Washington University

#### INTRODUCTION

During recent years, there has been a rising interest in the physiological, anatomical, and cytological study of the mosaic diseases of plants, as a result of the difficulty connected with a determination of the causal agency. Particularly has there been a cytological search of the tissues in an attempt to find either in the living or in the fixed and stained cell an organism which might prove to be associated with the disease. Notable among these studies have been those on tobacco by Iwanowski ('03), Hunger ('05), Delacroix ('06), Dickson ('22), Palm ('22), Goldstein ('24), Rawlins and Johnson ('24, '25), and Eckerson ('26); on potato by Smith ('24); on corn by Kunkel ('21); on the yellow stripe disease of sugar cane by Matz ('19); on the Fiji disease of sugar cane by Lyon ('10), Reinking ('21), and Kunkel ('24); on the mosaic and rosette of wheat by McKinney, Eckerson, and Webb ('23); and on Hippeastrum equestre Herb. by Kunkel ('22, '24), and McKinney, Eckerson and Webb ('24); and on Brassica pekinensis Skeels by Kunkel ('24).

In the course of these investigations various bodies and cell inclusions of different types have been described. Among other structures found in the cell have been the irregular, amoeboid-like, vacuolate or reticulate bodies which are in many ways comparable to the Negri bodies accompanying rabies, the Guarnieri bodies in small-pox, and the supposed Rickettsia micro-organisms of exanthematic typhus. These amoeboid bodies, as well as the other cell inclusions referred to, have been

<sup>1</sup> An investigation carried out at the Missouri Botanical Garden in the Graduate Laboratory of the Henry Shaw School of Botany of Washington University, and submitted as a thesis in partial fulfillment of the requirements for the degree of doctor of philosophy in the Henry Shaw School of Botany of Washington University.

shown to be usually connected with the chlorotic areas in the mosaic-diseased plants in which they occur, and many theories regarding their probable origin and their connection with the disease have been propounded. Hence, the purpose of this investigation was to study both fixed and living cells of numerous types of mosaics in order to determine of how general occurrence the bodies are in the cells of plants affected with mosaic diseases. It was hoped that, through a study of the living cells, some index as to the physiological nature of these bodies might be obtained.

Chlorosis, the pathological condition of which the mosaic disease is a type, has been defined by Clinton ('15) as "that unusual condition in a plant in which the chlorophyll loses its bright green color and becomes yellowish green or even white." The various types of chlorosis have been classified by the same author as follows:

- I. Infectious chlorosis.
  - A. Communicable through the juice.
  - B. Communicable through the tissues.
    - a. By buds.
    - b. By grafts.
- II. Non-infectious chlorosis.
  - A. Non-perpetuating.
    - a. Affecting plants generally.
    - b. Affecting isolated leaves or branches.
  - B. Perpetuating.
    - a. Through seed.
    - b. Through cuttings.
    - c. Through buds or grafts.

The mosaic diseases fall naturally in the class of infectious chloroses communicable through the juice. It was, therefore, of interest to make a cytological study of some of the other types of chloroses with the idea of determining how general an associate of chlorosis these various inclusions, and particularly the irregular vacuolate bodies, are. That is, the question arose as to whether these bodies were associated with the loss of chlorophyll in plant cells, or whether they accompanied only the infectious chloroses.

In the majority of the more common horticultural variegated varieties of plants, such as the crotens, oleander, Pittosporum, etc., chlorosis is non-infectious and is not communicable through either the plant juices or grafting. It is perpetuated in some cases through seeds; in some, through cuttings; and in still others through buds or grafts. However, Baur ('04, '06, '07, '08) showed that there were some variegations, particularly those in the *Malvaceae*, such as *Abutilon Thompsoni*, which were infectious in nature, since in grafting the variegated variety with the green the variegation could be transmitted either from stock to scion or vice versa. There has always been some feeling that Baur's infectious chloroses are not entirely unrelated to the mosaic diseases which are simply infectious chloroses transmitted through the diseased juice. Perhaps, when more is known of Baur's infectious chloroses, it will be found that there is some means other than grafting by which they also may be transmitted.

Histological studies of variegated leaves have been made with the idea of classifying them through their anatomical structure. The outstanding work along this line is that of Funaoka ('24). in which 18 different variegations were studied and classified on the basis of characters of their microscopic anatomy. None of these studies, however, gave any indication of the presence in the cells of bodies and inclusions comparable to those found in certain of the mosaic diseases. Hence, a cytological study of representatives of the various types of variegations was planned in order to determine whether there are any inclusions in the chlorotic areas comparable to those in the chlorotic areas of the mosaic diseases studied, and if so, whether they are in any way similar. It was hoped that such a study would give some indication of a possible relation between the infectious and noninfectious variegations and the mosaic diseases which are due to. and transmitted by, the filterable virus in the diseased juice.

# DISCUSSION OF LITERATURE

A mosaic disease was first carefully studied by Mayer ('86) in tobacco, but although he found that it could be transmitted by inoculation of the mosaic juice into healthy plants and that it was inactivated by heating to 80° C., he made no cytological study of the tissue. He did examine the sap but found only starch grains and calcium oxalate crystals. Koning ('99) drew

the cross-sections of a few leaves but failed to observe any pathological inclusions. It was Iwanowski ('03) who first made a detailed cytological study of the leaves of infected plants. studied living cells as well as cells of tissues which had been fixed in Flemming's solution, in osmic acid, and in boiling absolute alcohol, and of these he found the latter the most successful. In the living cells he observed a greater abundance of oxalate crystals in the chlorotic than in the green area; clear plate-like crystals which he described as being similar to waxy material, but less refractive; cells with granular bacteria-like inclusions which, however, never form uninterrupted complexes: and finally, plasma-like accumulations reminding one of parasitic amoebae. In the material fixed in boiling absolute alcohol and stained with methylene blue and eosin he found structures which he thought were zooglea of bacteria with approximately the same form as those of the plate-like crystals in the living cells. Also, he observed, as in the living cells, that the plasma accumulations took the eosin more deeply than did the cytoplasm, and these were most frequently found in the neighborhood of the nucleus or the crystals.

To these observations he gave the following interpretations: the plate-like crystals in the living material gave rise to the striate zooglea in fixed material, the latter being composed of organisms which were the causal agency of the mosaic and were small enough to pass through a filter; the amoeboid bodies, he believed, were the result of the reaction of the cell to the irritation of the causal agency.

Hunger ('05), in checking Iwanowski's work, showed that the so-called mosaic disease bacteria of Iwanowski, as well as the zooglea, disappeared when the cell was treated with phenol chlor-hydrate, while the other cell-structures remained the same. That is, he obtained a solution of the plate-like crystals with phenol chlor-hydrate.

Delacroix ('06), in following the method of Iwanowski of fixing the tissues in boiling absolute alcohol, was able to observe the amorphous bodies and the zooglea. However, when fixed in cold absolute alcohol he obtained no such structures. From these observations he concluded that they were deposits of

substances which were dissolved in the cell sap and precipitated by the boiling alcohol. He apparently did not account for the fact that they also appeared in the living cells. In view of this latter fact his results could no doubt be interpreted as having been brought about by the solution of the bodies in cold but not in hot absolute alcohol.

Lyon ('10), in studying the Fiji disease of sugar cane, found one or more plasmic bodies in every cell of the abnormal tissue. They were generally rounded in contour although they might be variously lobed or distorted at times, and their constituent substance was much denser optically than that of the cytoplasm of the cane, making them readily visible even though all the contents of the cells were clear and colorless. They were devoid of cell walls or other precipitated secretions. In interpreting these observations, he concluded that "In these foreign bodies we recognize an interesting and dangerous parasite quite new to the science of sugar cane pathology. It belongs to a very small group of lowly organisms whose position in the realm of living things is a subject of dispute among naturalists." ently he considered that they were plasmodia of a myxomycete, similar to Plasmodiophora Brassicae, which break up to form spores, the latter being freed with the disintegration of the tissue. Infection, then, according to him takes place by the entrance of the swarm spores through the roots and up through the vessels to the leaves.

Matz ('19), in his studies on the yellow stripe disease of sugar cane, observed granular, plasmodium-like substances in the yellow-striped cane leaf and stem tissues, and found the mass made up of small, hyaline bodies, the entire mass being in the form of a compact plasma with the bodies less than 1 μ in length. They seemed, however, less clearly defined than masses of bacteria. Later, in 1922, he considered them as co-generic with Strongyloplasma Iwanowski Palm, described by Palm in the chlorotic areas of the tobacco leaves infected with mosaic.

The term Strongyloplasma Iwanowskii was applied by Palm ('22) to the very small granules (at the most .5  $\mu$  in length) which he observed in the chlorotic areas of tobacco leaves infected with the mosaic disease. He found them staining black with haematoxylin, and frequently forming irregular agglomera-

tions which sometimes filled the entire cell lumen. These very small bodies he believed agreed in every respect with the Strongyloplasma of von Prowazek and Lipschütz. He also observed the fairly large corpuscles usually in contact with, or in close proximity to, the nucleus, staining gray with haematoxylin and very light red with eosin. In the living cells the latter were present and were denser and more opaque than the surrounding cell plasm. Although they were displaced at times by the streaming of the protoplasm, he saw no evidence of any automotive movement. In the interpretation of his results he agreed with Iwanowski ('03) that these latter bodies were the product of the reaction of the virus carrier on the cell plasm, and believed that they were homologous with the Guarnieri bodies. The very small granules were, according to him, the causal agency, and he therefore concluded that in the mosaic disease of tobacco we are dealing with a disease which belongs etiologically to the chlamydozoonoses.

The presence of the plasmodial bodies in the Fiji disease of sugar cane as described by Lyon ('20) was verified by Reinking ('21), who reported finding the bodies as light-colored in the younger galls and brown and granular in the older ones. He found them in the young shoots arising from the base of diseased plants, in the rotted roots, and in the base of the stem. He concluded that, although the presence of the bodies throughout the plant has not yet been demonstrated, their presence would indicate that "the fungus is responsible for the disease."

Kunkel ('24) studied the bodies in connection with the Fiji disease in great detail and found particularly in the young galls that they were frequently small and composed of a deeply staining granular mass with occasional vacuoles. In many cases, they possessed short blunt appendages which were more hyaline than the main part of the body and resembled pseudopodia of amoebae. In maturing and old galls, the bodies became larger and less dense, frequently containing large, deep-staining granules in a vacuole surrounded by a definite membrane. He frequently found two bodies, one in either end of the cell, in the case of dividing cells, thus indicating that these plasmodial bodies do possess the power of division and of growth. These facts,

together with the distribution of the bodies in the gall tissues, their apparent manner of spreading, and constant association with the earlier stages of gall formation, indicate, he believed, that they belonged to a parasitic organism. However, he still considered it an open question as to whether they were inert cell inclusions resulting from some physiological disturbance, or represented a new type of plant parasite.

Kunkel ('21) has also studied the mosaic disease of corn. and compared the bodies found there with those in the yellow stripe disease and the Fiji disease of sugar cane. Studies were made both of living and fixed material, and in all chlorotic areas irregular amoeboid bodies were found in association with the host cell nucleus and frequently appeared to be attached to it. They showed a structure similar to that of protoplasm but somewhat more dense and opaque. In the living cells, he never found them showing any automotive movements or change of form. Frequently they appeared naked, but at times a thin limiting membrane could be observed. Vacuoles were present in some and in others they were absent. Also, there were in some of the bodies numerous dark-staining granules which tended to be angular rather than spherical and never showed any semblance to nuclei. Those cells possessing the bodies were found to increase in size and to show nuclei which also were larger than normal. During the early stages of the disease the bodies were very minute and apparently increased in size with the progress of the disease. The outstanding characteristics, then, as he saw them, were that they appeared to grow, that they showed a structure like that of protoplasm, that they stained like protoplasm, and that they tended to be amoeboid in shape. Also. the tendency to cluster around the nucleus was comparable to the similar tendency among other known intracellular parasites such as the swarm spores of Chrysophlyctis endobiotica Schilb in the potato wart disease (Orton and Kern, '19). The bodies. therefore, in his estimation, had many of the characteristics of a living organism. However, they had never been cultured, and until that might be possible they could not be definitely considered as the causal agency.

The vacuolate bodies found here were entirely different from

the granular mass described by Matz ('19) in the yellow stripe disease of cane, since the latter did not stain like protoplasm, did not show a protoplasmic structure, were not vacuolate, did not contain any of the dark-staining granules, and were not plastic. Neither were they like the bodies associated with the Fiji disease of cane, nor like the plasmodia of *Plasmodiophora Brassicae*. They did, however, resemble remarkably the Negri bodies associated with rabies in the brain cells of the diseased dog.

Similar vacuolate bodies have been found in wheat plants infected with the rosette disease and the mosaic-like leaf mottling by McKinney, Eckerson, and Webb ('23a). They were found in the roots, crown tissue, leaf sheaths, and leaves, never necessarily occurring in close contact with the nucleus. Their contents were rather homogeneous in structure, and they contained many large and small vacuoles in which granules could be observed showing Brownian movement. No independent movement was ever observed in the living cells. From these observations they concluded that, although the bodies might be a stage of some definite parasite, yet their distribution in the host tissue and their apparent parallel development with that of the host cells did not seem to conform exactly with the distribution and development of any plant parasite known.

The same investigators (McKinney, Eckerson, and Webb, '23) have described similar bodies in the chlorotic areas of the leaves of Hippeastrum Johnsonii which were infected with the mosaic disease, as did also Kunkel ('24a) in Hippeastrum equestre Herb. Kunkel found them to be similar to those in corn as regards distribution, although the Hippeastrum bodies were considerably smaller. They were never found in tissues until the mosaic blotching appeared, and the size of the bodies was directly proportional to the degree of chlorosis, the lighter areas showing the largest bodies. Kunkel also found such amoeboid-like structures in the chlorotic areas of mosaic-diseased Brassica pekinensis (Skeels), which were similar to those in corn and in Hippeastrum equestre Herb, both in structure and in staining The Brassica bodies differed, however, in not being reactions. primarily associated with the nucleus. He then concluded, from all of his observations on the bodies in the Fiji disease of

sugar cane, in the mosaic disease of corn, in *Hippeastrum equestre* Herb., and in *Brassica pekinensis* (Skeels), that the amoeboid bodies associated with these diseases might represent only one stage in the life history of the causal organism, and that at another stage they might be so small and plastic that they could pass through the fine pores of a filter and escape detection under the microscope. If this were the case, they would probably become visible only after a certain period of growth within the cell of the host. For the time, however, he added, "we must be content with the knowledge that intracellular amoeboid organisms accompany the mosaic disease in several plants, that these bodies look like living organisms, and that in corn and *Hippeastrum* they are associated with chlorosis in such a way as to account for the mosaic pattern in the leaves."

Smith ('24) studied the leaf and stem tissue of varying age from mosaic-diseased potato plants and found in the chlorotic areas similar vacuolate bodies which showed definite walls and bore, as he said, "a superficial resemblance to some kind of protozoal organism." They were usually in close association with the nucleus of the host cell, and in the living cells they failed to show any automotive movement. In view of these facts and the further fact that in the light green areas in which these bodies were found the general disintegration of the tissue seemed to be considerable, he came to the conclusion that they were some type of degeneration product of the cell and probably of the nucleus, induced by the mosaic, and that they were the effects rather than the cause of the disease.

Contemporaneous with the studies on the mosaic disease and the associated amoeboid-like bodies in corn, sugar cane, potato, *Hippeastrum*, and *Brassica*, have been several notable contributions to a study of similar structures in the chlorotic areas of the mosaic disease in tobacco, tomato, and related genera. Dickson ('22), in observing living free-hand sections of the tobacco leaf, found in the chlorotic areas of leaves in the advanced stages of the disease, among the vacuolate bodies and the plate-like crystals, numerous smaller bodies exhibiting an erratic movement. He considered them as flagellates but in spite of careful staining he could obtain no proof of this. In fixed material he

found minute dark-staining bodies, .3 µ long and slightly less in width. They were found particularly in the border parenchyma of the vascular tissue of diseased leaves, but were also observed in close contact with the walls of the chlorenchyma cells and, in some cases, surrounding the chloroplasts. The vacuolate bodies, comparable to those described by Kunkel in corn, he believed were not the causal agency, but were, on the other hand, secondary in nature. He attempted to culture these very small bodies but was unsuccessful in isolating them from the virus. The plate-like crystals which have been described in connection with the mosaic disease of tobacco were believed by him to be due to a product of chlorophyll degeneration combined with changed plastid protoplasm, in view of the fact that Ewart had shown that CO2, combined with chlorophyll in the presence of water to form xanthophyll and a colorless waxy substance.

In connection with these studies Dickson also made an anatomical study of numerous mosaic-diseased plants, from which he drew the following conclusions regarding the anatomical characteristics of the mosaic disease in general:

1. There was a difference in thickness between the chlorotic and the green area, the ratio being about 2:3, due to hypoplasia of both palisade and spongy parenchyma cells in the chlorotic areas.

2. The dark green areas exhibited hyperplasia.

3. There was a regular arrangement of cells in the light area, thus reducing the intercellular spaces.

4. The epidermal cells were smaller in area but deeper over the

hypoplastic areas than over the normal.

5. Hypoplasia was accompanied by a degeneration of cell contents.

6. Disintegration of the chloroplasts was accompanied by the appearance of the very small hyaline bodies in rapid movement, the semi-crystalline plates believed to arise from degenerate chloroplasts, and the vacuolate bodies.

The vacuolate bodies in the hair cells of the mosaic-diseased tobacco were studied by Goldstein ('24) in the living condition. She found that in the chlorotic areas the hair cells and epidermal cells contained the vacuolate, more or less amoeboid bodies, and the crystals, both of which have been described and illustrated many times since their first description by Iwanowski ('03). However, she watched them and studied them more intensively

in the living condition than had been done heretofore. She found that the bodies bore no definite relation to the nucleus, but were simply in the cytoplasm and carried about in it when the protoplasm was streaming actively. Not only did she see the body carried around the cell in the protoplasmic streams but also she noticed an apparent change in form which might be described as an indication of automotive movement. In the pseudopodia of the more active bodies she occasionally observed a hyaline ectoplasmic-like cap bordered by a membrane. She interpreted her observations of such membranes as convincing evidence in favor of the view that the bodies are surrounded by a definite plasmatic membrane. Also, she found that treatment with acid caused the contents of the bodies to shrink, leaving visible the definite limiting membrane.

She mentioned the crystals but gave no results of her work on them, promising a paper in the near future. She did, however, treat the cells with various fixing fluids, watched the influence of these under the microscope, and found that the crystals lost their typical form, becoming long, irregularly lobed and striated masses which were stained deep yellow by the fixatives. This accounted for the appearance of the striate masses in the fixed mosaic tobacco tissues. She concluded from these observations that it might be possible that such plastic bodies as these would be able to pass through cell walls and the pores of bacterial filters just as the nuclei were observed to migrate from cell to cell in *Tradescantia* (Miehe, '01), thus explaining the nature of the virus.

Rawlins and Johnson ('24, '25) have studied cytologically the fixed tissue of the mosaic tobacco leaves, and have described three types of cellular inclusions,—the yellow-staining striate material radiating from the nucleus, small black-staining bodies, and vacuolate bodies varying in size from those just visible to those slightly larger than the nucleus. They found that the development of these inclusions was inhibited by temperatures which inhibited the expression of the mosaic. Also, it was observed that only 20 per cent of the plants showing the symptoms out of doors showed the bodies indoors, whereas 80 per cent of those in the greenhouse showed them. They have

attempted to show the sequence of the appearance of these various types of bodies. The first to appear was the striate material, the small dark-staining type, and the crescent-shaped vacuolate type, and they felt that the small bodies were a stage in the development of the vacuolate type. The crescent-shaped type definitely gave rise to the rounded vacuolate type which then persisted along with the striate material throughout the life of the leaf. They considered that the amorphous nature of the striate material indicated that it was a product of the diseased cell or of the causal agency.

Very recently Eckerson ('26) has described what she considered an organism of the tomato mosaic. She studied the tissues of tomato plants at various short intervals after inoculating the healthy plants and was able to observe at 24 hours after inoculation flagellate organisms in the veins and adjacent tissue, and the chloroplasts near the veins showed signs of dissolution. As the time after inoculation increased, the bodies became more numerous and the dissolution of the chloroplasts continued, while the bodies within them increased in size. Seven days after inoculation many of the chloroplasts in the palisade layer were in the process of liquefaction, while the remaining plastids contained non-motile bodies which seemed to be early stages of spore formation. Ten days after inoculation, when the leaflets began to show mottling, the palisade cells were partially disorganized, the cytoplasm was gone, the chloroplasts partially dissolved, and the remaining ones contained spores. These disorganized cells were usually bounded by groups of cells in apparently perfectly healthy condition. She has included many illustrations of both the spore and flagellate form, showing the nature of the organism. It is interesting that the organism should have been found associated with the chlorotic areas of mosaic-diseased plants, but she has not yet demonstrated that it is the causal agency by isolating it and inoculating the resultant culture into healthy plants. Moreover, it must be remembered that the size of the particle of the causal agency has been determined by Duggar and Armstrong ('23) through filtration experiments to be approximately the size of the particles in a fresh 1 per cent solution of haemoglobin, which is 30 uu. The smallest size which Eckerson

gave for the flagellate forms she described is 2-4  $\mu$ , and this is approximately 1000 times the size of the particle as determined by Duggar and Armstrong.

The work of Nelson ('23) has not been included in this discussion, since it has been completely refuted by other investigators who have shown that the protozoan-like bodies which he described occur normally in the phloem tubes of healthy plants.

Structures similar to Nelson's bodies have been described by Klebahn ('26) in the sieve tubes of Anemone nemorosa, and he believes that evidence is strongly indicative of their being the cause of the disease called by him "alloiophylly." He considers that these bodies are closely related to the bacteria in that they show no definitely organized nucleus, and that their size seems to fall within the limits of the size of bacteria; and he applies the term Scolecosoma anemones to them. They have not, however, been found to reproduce by simple fission, and for this reason as well as the fact that they exhibit a great variation in form, they differ from the bacteria. He has, therefore, concluded that they are a "neuen Organismengruppe die etwa zwischen Bacterien und Flagellaten vermittelt." According to him, it is possible that his Scolecosoma and Nelson's bodies belong to the same or nearly related species, and it should still be an open question as to whether or not these belong to the same group of organisms as do the vacuolate bodies described as accompanying many of the mosaic diseases.

There have also been numerous cytological and physiological studies made on the chloroses of many of our variegated horticultural varieties. Although anatomical studies have been made, particularly by Funaoka ('24), there have never been found inclusions of the nature of those described as occurring in several of the mosaic-infected plants. However, the investigations have led to results so closely parallel to those obtained from work on the mosaic plants that a resumé of them would be of interest here in showing the possible connection between the two types of chloroses.

Masters ('69) considered albinism as a change due to the deficient formation of green coloring matter or chlorophyll. He distinguished between this condition and etiolation by the

fact that in the former chlorophyll seemed never to be formed in the affected parts, even if they were exposed to light, while an etiolated organ placed under favorable circumstances speedily assumed a green color. Later, Weiss ('78) explained variegations on the assumption that white spots were caused by the presence of air which was held in the intercellular spaces under the epidermis. He showed that leaves which were exhausted of air under water, by means of a pneumatic pump, lost the white spots. However, Dalitsch ('86) contributed the first accurate observations on the cause of variegation. He defined the white spots as due to chlorophyll-free cells in the fundamental tissue.

Saposchnikoff ('89) studied the starch content of chlorotic and green areas of variegated leaves and found starch only where chlorophyll was present. When, however, the leaves were placed in sugar solution, starch was found in equal amounts in the chlorotic and the green areas. In this connection, Winkler ('98), from a study of leucoplasts, chromoplasts, and chloroplasts, concluded that, whether the stroma was or was not stratified, whether it contained chlorophyll or some other pigment, whether the pigment was granular or crystalline, whether the plastid was large, small, distorted, or smooth, the stroma was always, when not too greatly reduced, able to form starch, if sugar were present.

Pantanelli ('05) made a physiological study of the variegations and found that it was possible to explain them on the basis of enzyme action. He found that the chlorotic areas were characterized by a decrease in chlorophyll content, increase in the accumulation of oxidizing enzymes, an increase in the osmotic pressure, lack of accumulation of mineral and organic salts and sugars, and a limitation of growth processes. His explanation of these observations is given in the following theory regarding the etiology of variegations. The first indication was probably an abnormal accumulation of the oxidizing enzymes, which disturbance probably took place in the stem or root; and then the disturbing material was carried up by material transport through the sieve tubes to the various parts of the plant. In the chlorenchymatous cells, it led to a destruction of the chlorophyll and to a general disease of the protoplasmic parts, which was evidenced by an increase in turgor. Further investigation

showed that the protoplasm and the plastids were disorganized and digested by the enzymes which were developed in abnormal quantities. The observed increase in osmotic pressure, he believed, was probably due to the increase in disintegration products of smaller molecular dimensions. He therefore considered that the pattern of the variegation followed the veins, through which the agency was carried and from which it was frequently distributed on one side only, making the vein the boundary between the green and the chlorotic areas.

Baur ('04) distinguished between the non-infectious and the infectious types of variegation, the former being transmitted through the seed, whereas the latter, although they could not be transmitted through the seed, could be passed on to healthy plants by grafting a variegated twig on a healthy stock. found infectious variegation to be quite frequent among the Malvaceae, and he investigated in great detail the variegated variety Abutilon Thompsoni. A microscopic study of the leaf tissue revealed nothing of the nature of a causal organism, but only a reduction in the size and number of plastids and in the amount of chlorophyll contained by these in the chlorotic areas. In an attempt to determine the nature of the virus he tried many methods of transmitting it to healthy plants, but was successful only in the grafting experiments. In 1906, he concluded that the virus was not an organism but highly organized products of metabolism, which in a certain sense, possessed the power of growth. Such products passed through the cells of the plant, and in the embryonic cells of the young leaves they found free side-chains to which they attached themselves. In these cells, then, it was believed that the toxin was again formed anew. This physiological explanation is quite analogous to a similar explanation given by Hunger ('05) as the cause of the mosaic disease of tobacco.

A very similar theory was sponsored by Molisch ('08) in connection with his studies on *Abutilon Thompsoni*. He studied both living and stained material and found no structures which might be interpreted as living organisms. He also tried cultivating the virus on artificial media and on an extract of *Abutilon* leaves, but was entirely unsuccessful.

At this time, Kränzlin ('08) made a study of the pigments connected with variegated plants, and he obtained results which were confirmed later by the work of Colon ('19) in a study of the pigments associated with the mosaic diseases. These results indicate that the same pigments are present in both the chlorotic and the green areas, the difference being primarily one of quantity of pigment. In the chlorotic areas he found less of each of the normal leaf pigments, that is, he found a similar decrease in chlorophyll and in carotin. Colon, in his work, obtained similar absorption spectra from the two areas and concluded that the chlorotic nature of the spots in the yellow stripe disease of cane was not primarily due to a decomposition of the chlorophyll as such.

Küster ('19) found the veins incompletely developed in the chlorotic areas of the marginally variegated leaves of Acer platanoides, and hence concluded that degeneration or incomplete development of the green chloroplasts was probably caused by diminished nourishment. This was, however, refuted by Funaoka ('24), who studied the relative frequency of the veins in the chlorotic and the green areas of 14 species. The results showed that in 9 of the species there was an equal frequency, in 3 species there was a thicker network of veins in the chlorotic than in the green areas, whereas in a single species, Euphorbia marginata, the net was thicker in the green areas. In Richardia Elliottiana the vascular network was not developed in the chlorotic area. Hence, from his observations he concluded that in many plants the cause of variegation could not be traced back to an insufficient supply of vascular bundles and a resulting poor nutrition.

Funaoka also made an extensive study of 18 different variegations, and from his observations was able to classify them on the basis of their microscopic anatomical characters. The paper was well illustrated with semi-diagrammatic drawings showing that the chloroses might be due to a loss of chloroplasts in one or more layers of cells in the leaf (periclinal variegations), or to an anticlinal division of the green and white areas of the mesophyll, or a lack of differentiation of one or more layers, particularly the palisade layers. No indication of any structure resembling a microorganism was described.

From an investigation particularly of variegated varieties of Zebrina, Tsinen ('24) concluded that from the cytological point of view variegations occurred as the result of an alteration in the plastid mechanism of the cell. This alteration could take place before the differentiation of the plastids from the chondriosomes, at any stage during their development, or after they were mature, thus explaining the various types of variegations found.

### MATERIALS AND METHODS

Living and fixed materials were studied in plants infected with the mosaic disease and in the variegated plants. For a study of the living material the following vital stains were tried, —methylene blue, neutral red, bismarck brown, dahlia, and brilliant cresyl blue, the most favorable being the latter in concentrations of 1:10,000 and less. Thin sections of even the thinnest leaves, such as those of Bougainvillea, could be easily made by holding several pieces of the leaf within the same piece of pith, and then making free-hand sections of them. The sections were mounted in water and studied without any stain, after which a drop of brilliant cresyl blue solution, 1:10,000 was drawn under the cover-slip. The large vacuolate bodies, particularly those in Petunia, were found to stain very well after an exposure to the dye of from fifteen minutes to several hours.

For the fixed and stained materials, the following fixatives were tried,—Bouin's fluid (see Lee, '13, p. 65), Flemming's weak solution (see Chamberlain, '24, p. 25), medium chromacetic acid (see Chamberlain, '24, p. 25), osmic-sublimate mixture (see Lee, '13, p. 50), and an acetic-alcohol-formalin mixture (see Rawlins and Johnson, '25). In all cases small pieces of tissue were taken in order to insure rapid penetration.

These 5 different fixatives were tried on the tissue of tobacco, poke, and geranium mosaics, and on the variegations in *Evonymus japonica* Linn., *Ficus Parcellii* Veitch, *Ligustrum ovalifolium* Hassk., and *Abutilon pictum* Walp. in combination with each of the following stains,—Flemming's triple stain (see Chamberlain, '24, pp. 59-62), Delafield's haemotoxylin (see Chamberlain, '24,

pp. 45-48), and Haidenhain's iron alum haemotoxylin (see Chamberlain, '24, pp. 41-45).

The best results were quite generally obtained when the chrom-acetic acid was used as a fixative and was followed by the Haidenhain's iron alum haematoxylin. Counterstaining with the Orange G was found to be very desirable, since the vacuolate bodies seem to show a strong affinity for it. The most satisfactory method for introducing the counterstain was to dilute a 1 per cent solution of the stain in clove oil to a light amber color and place the slides in it for 10 to 15 seconds before placing them in xylol. This combination of chrom-acetic acid as a fixative, and Haidenhain's iron alum haematoxylin counterstained with Orange G was then used in all subsequent preparations, since it seemed the most generally successful.

With this technique, then, the following mosaic diseases were studied,—tobacco (Nicotiana Tabacum Linn.), petunia (Petunia sp.), columbine (Aquilegia caerulea James), pokeweed (Phytolacca decandra Linn.), and Jimson weed (Datura Stramonium Linn.). As to variegations, the following were chosen because they represent both Monocotyledons and Dicotyledons, because there are among them both infectious and non-infectious chloroses, and because they show quite different and distinct anatomical variations: Homalomena cordata Schott, Ficus Parcellii Veitch, Nerium Oleander Linn., Coleus Blumei Benth. var. "Mrs. Kirkpatrick," Bougainvillea glabra Choisy var. "variegata," Pittosporum Tobira Ait. var. "variegatum," Evonymus japonica Linn. var. argenteo-variegata, and E. japonica Linn. var. "mediopicta" Hort.

The cytological studies were made with a Zeiss microscope equipped with a 1/12a fluorite or semi-apochromatic objective, and a 4-mm. achromatic objective number 6. While studying the preparations the binocular tube was used with number 4 oculars, but all drawings, with the exception of text-figs. 2, 3, and 4, were made with the monocular tube, using a 12x compensating ocular. The text figures just mentioned were drawn with a 2x ocular and the 4-mm. objective. All drawings were made with the aid of a Spencer camera lucida.

# OBSERVATIONS AND DISCUSSION MOSAICS

Preliminary to the following work, a survey was made of living and fixed material of many of the mosaics at the author's disposal, with the idea of determining those which seemed most favorable for more intensive study. Several which are not mentioned here, such as *Crotalaria*, geranium, and poinsettia, have not as yet been sufficiently studied to warrant a report in the present paper, but it is hoped that the work on these may be forthcoming in the future. The studies given here have been made upon tobacco, *Petunia*, *Datura*, pokeweed (*Phytolacca decandra*), and *Aquilegia caerulea*.

1. Tobacco.—The work began with a study of the hair cells in the chlorotic areas of tobacco, since the inclusions here have been frequently described and since these cells seemed to offer such good material for the study of the bodies and crystals in living cells. Epidermal and hair cells were studied in the living condition, while in the case of fixed materials cross-sections of the leaves were used. In the living cells, irregular, vacuolate, amoeboid-like bodies together with clear plate-like crystals were found just as illustrated by Goldstein ('24). No indication of anything comparable to a nucleate structure was observed in either the fixed or living bodies, and they appeared to lack a limiting membrane of any sort. In the living cells, when protoplasmic streaming was sufficiently rapid both the bodies and the polygonal, flat, plate-like crystals were carried with it around the cell, but at no time could any movements which might be interpreted as automotive be discerned. The vacuolate bodies apparently changed their form somewhat, but all of these changes could be explained by the fact that the body was being turned over in the stream just as were the large crystals. There seemed to be no connection between the nucleus and the vacuolate bodies, the only times when they were adjacent to each other being when the nucleus acted as an impediment to the body as the latter was being carried through the cell in the protoplasmic Both the bodies and the crystals were strikingly similar to those in Petunia, and since they will be described later, it is not necessary to go into detail at this point.

Rawlins and Johnson ('25) showed that in tobacco mosaic the bodies were found in 80 per cent of the mosaic plants grown under greenhouse conditions, whereas only 20 per cent of those grown outside exhibited them. In the present investigation it was also found that in the tobacco the inclusions were of more common occurrence in the greenhouse plants than in those grown out of doors. The idea arose that perhaps the ultraviolet rays which reach the plant when it is grown outside but which are cut out by the glass of the greenhouse might be, at least in part, the cause of this difference. With this idea in mind the following experiments were conducted.

# EFFECT OF THE LONGER ULTRA-VIOLET RAYS ON TOBACCO PLANTS INFECTED WITH MOSAIC DISEASE

Ultra-violet rays may be divided into two diametrically opposed categories in regard to the action on living organisms. Those with the longer wave-lengths, 400-290 µµ (4900 A. U.-2900 A. U.) are commonly known as the biological rays. Since they are relatively penetrating, they include that range of the ultra-violet which may be present in the solar spectrum as it reaches the earth's surface. As to their action on living organisms, they are characterized by being chemically oxidizing and hence metabolic synergists. Opposed to this division, are those with the shorter wave-lengths which are commonly termed abiotic rays, because of their action on living protoplasm. In contrast with the longer rays, they are chemically reducing and metabolic depressors. They are so readily absorbed that penetration is very slight, hence they are never present in the solar spectrum as it reaches the earth's surface. In fact, the shortest wave-lengths obtained in the solar spectrum are about 291 uu. All the shorter wave-lengths are absorbed by the earth's atmosphere. Because of the poor powers of penetration the abiotic rays are known to be superficial in action, being unable to penetrate the human epidermis. It is, then, recognized that the abiotic rays are lethal to bacteria and other living organisms. and these are concerned in sterilization processes.

Since the abiotic rays are so poorly penetrating and since it is the biological rays which have such profound reactions on the tissues of higher animals, it was the effect of these latter rays on the tobacco plants infected with mosaic in which the author was primarily interested. In the experiments 2 types of lamps were used,—the Alpine Lamp, of the Hanovia Chemical Co., and the Air-cooled Quartz Mercury Vapor Lamp, of the Burdick Cabinet Co. Both the biological and the abiotic rays can be obtained from these lamps, depending on the distance between the burner and the object exposed. A column of air of 36-40 inches will absorb the abiotic rays, leaving only the longer biological rays. Therefore, to test the effect of the abiotic rays, the object is placed within 6 inches of the burner, whereas in an investigation of the longer wave-lengths, the object is placed at least 36-40 inches from the burner.

In the literature there has been very little experimental work on the effect of the biological ultra-violet rays on the tissues of plants, hence many difficulties arose in connection with the details of applying the lamp. The plants were found to burn most easily, particularly with the new Burdick lamp, so the time of exposure and the working distance (i. e., the distance between the burner and the plant) had to be determined for each lamp.

The best results were obtained with the Hanovia lamp, because it contained a very old burner in which the intensity of the rays had been decreased considerably. With this lamp it was found that the plants could be given a treatment of 30 minutes at 40 inches daily without bringing about fatal injuries to the plants, although they did become severely dwarfed.

Six plants were inoculated with filtered juice from the leaves of mosaic-infected tobacco plants, and the ultra-violet treatment was begun 2 days later, after the plants had recovered from the ill effects of the inoculations. They were rayed daily for 30 minutes at 40 inches, being kept, during the remainder of the day, under normal greenhouse conditions. At the end of 9 days all 6 plants showed the normal mosaic symptoms. With continued treatments the plants gradually became more and more dwarfed, and with the noticeable dwarfing the symptoms became less evident. At 20 days after inoculation the plants had apparently completely lost all of the typical mosaic symptoms. The treatment was continued for an additional 8 days, thus making the entire treatment 4 weeks in duration.

After treatments were stopped the plants were kept under observation in the greenhouse for 40 days, at the end of which time 2 of the 6 plants had re-developed the mosaic symptoms, 2 remained uniformly green, and the remaining 2 succumbed to the treatment which they had received. Therefore, the loss of symptoms after 18 days of exposure to the ultra-violet was apparently simply a masking of them, such as has been observed in plants which have been kept under blue light (Lodewijks, '10; Chapman, '17; and Dickson, '22). There was not a permanent curative effect. As has been noted, the plants were dwarfed, showing that the conditions to which they were subjected during treatment were not optimum for growth, and it is a well-known fact that the symptoms are the most prominent when the plants are growing rapidly. Therefore, it was not surprising that the disease was masked.

This masking of the mosaic symptoms in tobacco plants which were exposed to the biological ultra-violet rays would seem to substantiate the work of MacMillan ('23), in which he observed a masking of the mosaic symptoms in potato plants grown at high altitudes. He suggested at that time that the masking might be due to the biological ultra-violet rays which are more abundant at high than at low altitudes, and which, he believed, stimulated chlorophyll production in these cells which would be chlorotic under ordinary circumstances.

Pieces of the leaves were taken at random from the plants when the treatment was completed, and studied in fixed material as well as in the living condition. The vacuolate bodies and the crystals were still present in some of the hair cells, indicating that the ultra-violet rays were not the cause of the absence of the bodies in the mosaic plants which were grown out of doors. Little effect was found in the tissues other than a more uniform distribution of the plastids and a more constant differentiation of palisade and spongy mesophyll throughout the leaf of the rayed mosaic plants; likewise, in the latter there was an increase in the number of epidermal hairs over that in the control plants.

Experiments conducted in the same manner with the Burdick lamp gave comparable results, but in the case of this new burner the intensity of the rays was so great that the burning and injury to the plants were difficult to avoid, even with exposures of only 1 or 2 minutes at a working distance of 50 inches. This again illustrates the great difference between burners and shows that the time and working distance of exposure must be determined for each burner with each type of plant. From these results it can be concluded that, in the case of the tobacco plants infected with mosaic, the only effect of the biological ultra-violet rays is the inhibition of normal growth, which in turn causes a masking of the mosaic symptoms. Moreover, the fewer inclusions in the cells of the plants grown out of doors than in the greenhouse plants cannot be explained as a result of the action of the biological rays of ultra-violet light.

#### EFFECT OF THE SHORTER ULTRA-VIOLET RAYS ON THE MOSAIC VIRUS

This study of the effect of the biological ultra-violet rays on the tissues of plants infected with mosaic suggested the idea of determining the effect of the abiotic rays on the virus itself. As has been explained, the shorter wave lengths are not penetrating and can be obtained only at the very short working distance of 6 inches. Hence, the effect of the abiotic rays on the virus cannot be studied by subjecting the plants directly to these for two reasons: first, the shorter wave-lengths are so poorly penetrating that they cannot pass through the epidermal cells; and, second, the plants so treated would be immediately burned and killed. In these studies, therefore, the effect of the rays on the virulence of the mosaic tobacco juice was investigated.

Fresh mosaic tobacco leaves were ground, the juice filtered through cotton, water added to make a 1:4 dilution, and this filtered through a spherical atmometer cup. This procedure removed from the juice the chlorophyll, which, if it had been present during the subsequent treatment, would probably have absorbed all of the shorter wave-lengths and a determination of their effect on the virus would have been impossible. The filtered juice, however, was a clear solution, thus avoiding the suggested difficulty.

Five-cc. portions of this clear, filtered mosaic juice were placed in uncovered 50-mm. petri dishes and then exposed to the ultraviolet rays at a distance of 6 inches for varying periods of time as seemed desirable from the preliminary experiments. Ten plants were then inoculated with the rayed juice in the case of each exposure, and, as a control, 10 plants were inoculated with the filtered juice which had not been rayed. The experiments were run at 2 different times during the winter and in different compartments of the greenhouse, with similar results, thereby assuring that environmental factors were neither favoring nor masking the expression of the symptoms. The virulence of the virus was considered to be indicated by the percentage of plants showing the mosaic symptoms. Since preliminary experiments showed that the virus was in no way inactivated by an exposure of 5 minutes, exposures shorter than that were not repeated in these two series of experiments. In this work, the new Burdick lamp was used, operating at 8 amperes and 70 volts. The results are given in the following tables.

TABLE I
INFLUENCE OF ABIOTIC RAYS ON THE MOSAIC VIRUS

Length of Exposure	Results								
Lengui of Exposure	1st series	2nd series							
Control 5 minutes 10 minutes 20 minutes 30 minutes	9 affected in 9 days 8 affected in 9 days 5 affected in 9 days 1 affected in 9 days None affected in 9 days	10 affected in 11 days 10 affected in 12 days 6 affected in 14 days 2 affected in 14 days 1 affected in 14 days							

In the experiments the remainder of the plants continued healthy for a period of 5 weeks and were discarded at that time. These results show clearly that the virus has, with sufficient exposure to the abiotic rays, been permanently inactivated. The same data show that the process of inactivation has been gradual, since there is some reduction in the virulence of the virus with an exposure of 10 minutes. The exposure is not lethal, however, until the virus has been subjected to the rays for a period of 30 minutes. The single plant which succumbed to the disease in the exposure of 30 minutes in the second series may have been an accidental infection.

The nature of this inactivation is not understood. An explanation of this process would probably at least suggest the

nature of the causal agency. The inactivation is not of the same nature as the killing of bacteria by the abiotic rays, since the time required is of an entirely different order. In the inactivation of the mosaic virus, exposures as long as 30 minutes are necessary, whereas, so far as known, all micro-organisms are killed by exposures which are measured in seconds rather than minutes.

The writer tried the influence of the rays from this same burner on  $Bacillus\ prodigiosus$ , with the idea of comparing the killing time here with the inactivation time in the case of the virus. Transfers of the organism were made, and when the cultures were 48 hours old a suspension was made in sterile distilled water. Plates were poured which contained 1 cc. each of a 1:10,000 and 1:100,000 dilution of the original suspension, in order to determine the density of the suspension. Counts of these plates showed that the original suspension contained approximately  $12\times10^5$  organisms per cc.

The original suspension and a 1:1,000 dilution of that suspension were then treated by the same method as was the virus in the above experiment, 5 cc. of the suspension being removed with a sterile pipette and placed in sterile petri dishes which were kept covered except during the short periods of exposure. Duplicate exposures were made in each case, and two plates inoculated from each exposure. The plates each contained a 1-cc. sample from the 5-cc. portion exposed to the rays. The results obtained are tabulated in table II.

TABLE II

EFFECT OF ABIOTIC RAYS ON BACILLUS PRODIGIOSUS

		Exposure										
	½ n	ninute	1 minute									
	Series A	Series B	Series A	Series B								
Orig. suspension	6 2	7 0	5 2	2								
1:1,000 dilution	0	0	0 0	0								

These results show that a suspension of Bacillus prodigiosus

of 12 × 10<sup>5</sup> per cc. can be practically killed by an exposure to the shorter ultra-violet rays given by the Burdick lamp when operated at 70 volts and 8 amperes. The killing time, which is 30 seconds in the case of *Bacillus prodigiosus*, therefore, is certainly not of the same order as the time of exposure required for inactivation of the virus. That this short killing time is a common characteristic of many organisms is shown in fig. 57 of Ellis and Wells ('25) which is included here as table III.

TABLE III

TIME IN SECONDS REQUIRED TO DESTROY VARIOUS ORGANISMS AT A
DISTANCE OF 200 MM. FROM A QUARTZ MERCURY LAMP
OPERATING AT 66 VOLTS AND 3.5 AMPERES

Staphylococcus aureus		- -	_ _	_					_	_			_	_	_	_
Vibrio cholerae	.	-	_ -	_								_		_	.	
B. dysenteriae (Shiga)		-	_ -	_		_				_				_		
B. dysenteriae (Dopter).	.	-	_ _	_		_		_						_	_	
B. coli	.	- -	_ -	_		_				<u> </u> .						_
B. anthracis (Sporogenus	)	+	_ _	_		_										
Pneumobacillus (Fried).		+	_ -	_		_									_	
Sarcina alba	. -	+	-	_		_	_							_	_	
Aerogenes capsulatus	.	+	_ _	_					<u> </u>		_					
B. tetani	.ــــ	4	_	_		_		_		_	_					
B. megatherium	.	╄	+	_												
B. phleole	.	+	+	_							_					
B. subtilis	.	╄	4	_				_								
Sarcina lutea	.⊢	╀	+	_									_		_	
Paramoecia	.	╀	+	-				_								
Yeast		╀	4	+	_					_			-	_	_	_
Time in Seconds	o i	20	40	60	.8	10	0 12	30 14	10 16	0 18	0 20	0 22	0 24	B	30 2	0 8

It is of interest to note from the table just referred to that the spores are not much more resistant to the abiotic effect of the rays than is the vegetative growth. According to Ellis and Wells von Recklinghausen has shown that while spores are 20 times more resistant than the vegetative forms to the action of chemical germicides, they are only 3 times more resistant to the ultraviolet rays than the vegetative forms.

These experiments show, then, that the abiotic rays can inactivate the virus if the latter is exposed to them for a sufficient length of time, but this time factor is many times greater than that which is necessary for the killing of the common micro-

organisms, either in spore or vegetative form. Hence, these results would seem to indicate that the virus is not an organism in nature. Whether the time factor is comparable to that which is necessary for the inactivation of enzymes has not yet been determined, and such determinations will give further indication as to the nature of the virus. The inactivation may simply have been due to a precipitation of certain proteins, since with the longer exposures a certain turbidity was observed in the formerly clear solution, and it is known that the rays will decrease the stability of the solution of some of the proteins, particularly the albumens.

2. Petunia sp.—A study of living free-hand sections of leaf tissues of healthy petunia plants and those infected with mosaic revealed in the latter the vacuolate bodies and the clear plate-like crystals similar to those observed so frequently in tobacco as studied particularly by Goldstein ('24). Just as in tobacco, the hair cells and the epidermal cells offered unusually favorable material in which to study the bodies in the living condition.

Although several leaves were studied from each of numerous healthy plants, no such inclusions were found. The normal petunia hair cell, as shown in pl. 13, fig. 1, contained only the nucleus, the cytoplasmic threads, and occasional small plastids carried along in the streams. The nucleus was found either suspended in the vacuole of the cell by the cytoplasmic threads, or closely pressed against the edge of the cell.

In the leaves of plants infected with mosaic, the hair cells in the dark green areas were similar to the healthy cells, showing no unusual inclusions. In the chlorotic areas, however, there were universally present both the vacuolate bodies and the plate-like crystals. Contrary to the distribution found by Goldstein in tobacco, there were never more than one or two of the vacuolate bodies present in a single cell at a given time. She illustrated as many as five in a single hair cell. The crystals, however, were present sometimes singly (pl. 13, fig. 2); sometimes as two or more separate and distinct crystals, each being carried about in the streams by itself; and sometimes in masses of numerous individuals lying adjacent to each other but not fused (pl. 13, fig. 19).

The cells were studied unstained as well as treated with various vital stains. The most successful vital stain employed was brilliant cresyl blue. The cells were mounted in water, and after they had been studied in the unstained condition a drop of a 1:10,000 solution of the stain was drawn in under the cover slip. The stain was taken up in from 15 minutes to 2 hours by the bodies, which could then be identified in the mesophyll, as well as in the epidermal hair cells.

The bodies exhibited different forms, varying from more or less definitely rounded, finely or coarsely granular structures, to irregular, vacuolate, amoeboid-like bodies. A limiting membrane was never observed except when the cells had been treated with 15 per cent alcohol and shrinkage had taken place, leaving visible the structure which was apparently a limiting membrane, as shown in pl. 13, fig. 9. The vacuoles varied in size. some bodies containing several large ones, and others numerous smaller ones giving them a porous or spongy appearance. They were not definitely associated with the nucleus as Künkel ('21) described them in corn, although at times they did appear adjacent to it. However, when the living cells were watched for a considerable length of time it was found that in those which were actively streaming the bodies were carried along in the streams. The nucleus frequently acted as an impediment in the course of the body, delaying its passage through the cell. If the cell had been observed only at that particular time the natural conclusion would have been that there was some association between the nucleus and the body, whereas a continued observation of the same cell showed that this was not the case. was one of these bodies seen in the process of division.

Single cells were watched for 2- and 3-hour periods of time, and the bodies and crystals observed as they were carried along in the protoplasmic streams. During such observations over long intervals of time, the bodies were seen to change in shape as well as to advance with the streams. The cell illustrated in pl. 13, fig. 2, was kept under observation for 2 hours, and during that time the body was perpetually changing form, as shown in the 40 camera-lucida sketches in text-fig. 1. The body apparently sent out short pseudopodium-like projections and these pro-

jections always appeared in the direction in which the body was being carried by the protoplasmic streaming. When meeting an obstacle such as the crystal, the nucleus, or some of the plastids, it would be stopped for a short time, then shape itself around the obstacle, and in a short time pass around it and continue to be carried in the stream. When reaching the end of the cell, the body could frequently be seen to flatten out in the stream against the cell wall and then again round up and continue its course back through the cell.

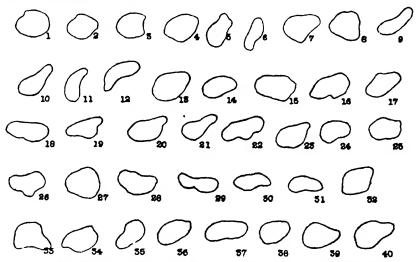


Fig. 1. Camera-lucida sketches showing changes in outline exhibited by the vacuolate body in the diseased hair cell of Petunia shown in pl. 13, fig. 2. These changes occurred during a two-hour period of observation while the body was being carried through the cell by the rapidly streaming protoplasm.  $\times$  1250.

At first, these movements were interpreted as being automotive, but further study seemed to show that they probably were not. If they were automotive, why did such movements occur only in those bodies in cells which were showing rapid streaming? They were never witnessed in cells unless there was a very pronounced cyclosis. Similar movements were observed by Goldstein ('24) in the bodies in the hair cells of tobacco and have been interpreted as sluggish amoeboid movements, but the author does not agree with her interpretation. If they were autonomous movements they should occur in the trichome cells independently of the streaming movements of the cyto-

plasm. Until further investigations have been conducted, such as attempting to remove the bodies from the cells by micro-dissection, it is impossible to state whether or not these bodies have automotive powers. At present, however, the author favors the view that the bodies are not the causal agency, but rather the result of a reaction between the cytoplasm and the virus. The apparent autonomous movements may have one of the 3 following explanations, or a combination of them. In the first place, these apparent changes may be due to the rolling over of the body in the moving stream of particles; secondly, the particles may be so mobile that the pressure of the cytoplasm as it pushes this larger object around the cell may be sufficient to cause changes in form, and finally, perhaps, to surface tension changes. The true explanation is probably a combination of the three.

Similar bodies are found in the fixed material, some of them being rounded and more or less definite, as in pl. 13, fig. 7, while others are more irregular and amoeboid in appearance (pl. 13, fig. 8). These are similar in appearance to those described by Iwanowski ('03), Palm ('22), Rawlins and Johnson ('24, '25), and Goldstein ('24). That they are the same structures which were studied in the living cells, I have demonstrated by treating the living cells with chrom-acetic acid under the microscope. Thus treated the fixed structures were identical with the bodies in the living cells.

The plate-like, highly refractive crystals were also studied. These, too, were observed to be transported in the cytoplasmic streams, but usually not as rapidly as the smaller bodies. As they were turned over by the pressure of the rapidly streaming cytoplasm, they showed various appearances, sometimes appearing long and narrow when viewed on edge; often appearing almost ribbon-like when viewed at an angle; and at other times as the flat plate-like structures shown in pl. 13, figs. 2 and 19. Their nature has not been determined, although the effect upon them of various solvents has been tried. They are definitely associated with the chlorotic areas of the leaves of plants infected with mosaic. They are not the result of a precipitation of products in the cell-sap, because they are definitely carried in the cyto-

plasmic streams. When studied in the living cells of tobacco by Dickson ('22) they were interpreted as being the product of chlorophyll degeneration combined with changed plastid protoplasm. His interpretation was based on the suggestion of Ewart's that CO<sub>2</sub> combined with chlorophyll in the presence of water to form xanthophyll and a colorless waxy substance. It seems improbable, however, that this is the explanation of the crystals obtained here, since in the epidermal and trichome cells in which they were studied the chloroplasts were few in number and very small in both the healthy and mosaic plants.

In the fixed material the crystals were not well preserved but were either striated in appearance or completely dissolved. These striated structures were observed by Iwanowski ('03) and interpreted as being plates of bacteria which were not visible except as the clear plate-like crystals in the living tobacco cells. As yet, no satisfactory explanation has been given of them, and there is none to be offered at present.

With the view of getting some indication of the nature of the bodies and the crystals in petunia, the solvent action of various substances, such as alcohol, formalin, KOH, HCl, HNO<sub>3</sub>, and CH<sub>3</sub>COOH were tried with the following results.

# EFFECT OF ALCOHOL ON THE INCLUSIONS FOUND IN THE PETUNIA LEAVES INFECTED WITH MOSAIC

Free-hand sections of the chlorotic areas were made and studied in water mounts, thus insuring the presence of bodies and crystals. The sections were then placed in small vials containing approximately 1 cc. of the solvents, and examined at more or less frequent intervals during a 12-hour period. The solubility of the vacuolate bodies in the various dilutions was entirely different from that of the crystals, both products, however, being soluble in 95 per cent alcohol.

These results show that the bodies are soluble only in 95 per cent ethyl alcohol, whereas the crystals are attacked in as low as 10 per cent dilutions. Plate 13, figs. 9-12 inclusive, show bodies in cells which have been in 15, 30, 50 and 70 per cent alcohol for 12-hour periods, showing that they are not dissolved at those concentrations. When attacked by the alcohol, the crystals were seen to swell and eventually burst and become uniformly

EFFECT OF DILUTION OF ALCOHOL ON SOLUBILITY OF INCLUSIONS			
Solvent	Bodies	Crystals	
C <sub>2</sub> H <sub>4</sub> OH 5% C <sub>2</sub> H <sub>4</sub> OH 10% C <sub>2</sub> H <sub>5</sub> OH 15% C <sub>2</sub> H <sub>5</sub> OH 30% C <sub>2</sub> H <sub>5</sub> OH 50% C <sub>2</sub> H <sub>5</sub> OH 70% C <sub>2</sub> H <sub>5</sub> OH 95%	Not affected Not affected Slightly plasmolyzed Not affected Not affected Not affected Dissolved	Not affected in 12 hrs. Dissolved after 6 hrs. Dissolved immediately Dissolved Dissolved Dissolved Dissolved Dissolved	

TABLE IV

distributed throughout the cell. This swelling took place immediately in all dilutions down to 10 per cent, and in this latter case the crystals were not completely dissolved until they had been kept in the solution for 6 hours.

EFFECT OF FORMALIN ON THE INCLUSIONS FOUND IN PETUNIA LEAVES INFECTED WITH MOSAIC DISEASE

Both the bodies and the crystals were found to be fairly resistant to the action of formaldehyde in 4, 8, and 12 per cent concentrations, the following results being obtained.

TABLE V SOLVENT ACTION OF VARYING CONCENTRATIONS OF FORMALIN ON INCLUSIONS

Solvent	Bodies	Crystals
Formalin 4%	Not affected	Not affected immediately, but some disintegration after 12 hrs.
Formalin $8\%$ Formalin $12\%$	Not affected Some disintegration	Show signs of disintegration Some disintegration

The disintegration accompanying the treatment of the cells with 12 per cent formalin was not specific for the inclusions alone, since there was also a general disintegration of the cell contents. Plate 13, fig. 19, shows a portion of a cell containing a mass of crystals and a single body which had been in 4 per cent formaldehyde for 1 hour. All of the inclusions were still perfectly normal in appearance, showing a resistance to the solvent action of formalin. After having been exposed to the same solvent for 12 hours, however, the crystals began to exhibit the effects of the solvent action, losing their angular corners, becoming rounded off, and occasionally showing signs of the dissolution of the crystal. as in pl. 13, fig. 18. Results comparable to these were obtained at the end of 1 hour with 8 per cent formalin, as is shown in pl. 14, fig. 33. On the whole, however, one may conclude that the bodies and the crystals are relatively resistant to the solvent or disintegrating action of formaldehyde.

# EFFECT OF ACIDS AND ALKALIS ON THE INCLUSIONS FOUND IN PETUNIA LEAVES INFECTED WITH MOSAIC DISEASE

To determine the effect of an alkali, KOH was used in concentrations ranging from .25 to 4 per cent. In all cases all of the inclusions were attacked immediately, the cell contents being rendered almost homogeneous, except in the case of the .25 per cent solution. In this dilute concentration dissolution was not immediate, but gradual. All of the inclusions, however, disappeared within 6 hours.

Diametrically opposite results were obtained regarding the solvent action of the mineral acids, HCl and HNO<sub>3</sub>. With concentrations as high as 10 per cent neither the bodies nor the crystals were injured, although the cells were severely plasmolyzed, as is shown in pl. 14, fig. 34. The results with acetic acid, however, were entirely different, the bodies not being injured by any concentration up to 10 per cent, whereas the crystals were dissolved in 20 minutes in all concentrations above 1 per cent, and in that concentration they were also dissolved in 1½ hours. These results of the effect of acids have been tabulated in table vi.

TABLE VI SOLVENT ACTION OF ACIDS ON INCLUSIONS

Solvent	Bodies	Crystals
HCl 5% HCl 10% HNO <sub>1</sub> 10%	Not affected Not affected Not affected	Not affected Not affected Not affected
CH,COOH 1% CH,COOH 2% CH,COOH 4% CH,COOH 5%	Not affected Not affected Not affected Not affected	Dissolved in 1 hour Dissolved in 20 minutes Dissolved in 20 minutes Dissolved in 20 minutes
CH <sub>1</sub> COOH 10%	Not affected	Dissolved in 20 minutes

From these studies, it can be concluded that the vacuolate bodies resemble the crystals in being relatively resistant to formalin, HCl, and HNO<sub>3</sub>, and soluble even in .25 per cent

KOH. They differ, however, in their solubilities in alcohol and acetic acid, the crystals being dissolved in 10-95 per cent concentrations of alcohol and in 1-10 per cent concentrations of acetic acid, whereas the bodies are soluble only in 95 per cent alcohol and never in acetic acid. It is hoped that further studies in this direction and in micro-dissection may reveal something more fundamental regarding the nature of both the bodies and the crystals, together with an explanation of their connection with the chlorotic areas in mosaic-infected petunia and tobacco plants.

3. Datura Stramonium Linn.—Healthy leaves of Datura, together with those of plants infected with mosaic, were studied in material which was fixed in chrom-acetic acid and stained with Haidenhain's iron alum haematoxylin and Orange G. Leaves of approximately the same age were taken from the healthy and the mosaic-infected plants from which the histological and cytological studies here reported were made. It was found in the histological studies that neither the green nor the chlorotic areas of the mosaic-infected plants are comparable to the normal, the green being thicker than the healthy but showing normal tissue differentiation and distribution of plastids; whereas the chlorotic areas are approximately equal in thickness to those of the healthy but have poorly differentiated palisade tissue and show a decrease in the number of plastids. These differences are brought out in text-fig. 2, and the measurements are given in table vii. The figures given in table VII are in each case the average of 50 measurements made on numerous sections of different pieces of fixed material.

TABLE VII

COMPARATIVE MEASUREMENTS OF TISSUES IN HEALTHY AND MOSAIC
INFECTED DATURA

	Healthy	Green area of mosaic	Chlorotic area of mosaic
Thickness of leaf Thickness of upper epidermis Thickness of palisade Thickness of spongy mesophyll Thickness of lower epidermis	107.2 µ	208.9 µ	124.7 µ
	10. µ	15.8 µ	19.4 µ
	48.9 µ	96.0 µ	40.3 µ
	40.3 µ	75.6 µ	48.1 µ
	10. µ	19.4 µ	17.1 µ
No. of rows of palisade cells	1.	1.	1.
No. of rows of spongy mesophyll	4–5	5-6	3-4
Longest diameter of chloroplasts	4.4 μ	5. µ	4. μ

A comparison of the measurements for the dark green area with those of the chlorotic area shows that the thickness of the leaf in the dark green area is 1.68 times as great as that in the chlorotic region; the palisade tissue is 2.38 times as thick; and the spongy mesophyll only 1.56 times as deep. This shows that the difference between the dark green and the chlorotic areas of the mosaic-infected plants is greatest in the palisade layer.

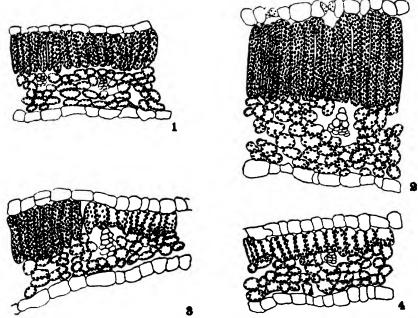


Fig. 2. Datura Stramonium. Semi-diagrammatic camera-lucida drawings of cross-sections of leaves, showing the effect of the mosaic on the tissues. × 300.

1, healthy leaf; 2, dark green area of a mosaic-infected leaf; 3, transitional area between the chlorotic and dark green areas of a mosaic-infected leaf; 4, chlorotic area of a mosaic-infected leaf.

This poor differentiation is shown more clearly in figs. 2, 3 and 4, of text-fig. 2.

On the other hand, a comparison of the measurements for the dark green areas of the mosaic-infected plants with those for the normal leaves shows that the increase in size of the dark green mosaic tissue over that of the healthy is approximately equal for all parts of the leaf. The thickness of the mosaic leaf in the dark green region is 1.96 times that of the healthy leaf; the upper epidermal cells are 1.94 times as deep; the palisade layer is 1.96 times as high; the mesophyll layer is 1.88 times as thick; and the lower epidermis is 1.58 times as thick. Hence the dark green area, although exhibiting an enhanced thickness, shows no abnormal tissue differentiation. The plastid distribution, also, is similar in the dark green area of the mosaic-infected leaves and the healthy tissue.

In the diseased leaves, the transitional areas between the chlorotic and the dark green regions are very sharp, and apparently follow the veins closely, as shown in fig. 4 of text-fig. 2. In one case, there was a dark green area existing between 2 veins which were only 350  $\mu$  apart. The dark green area which measured 205  $\mu$  in the widest reace dropped to 115  $\mu$  beyond either of the two limiting veins. Thus, in the short distance of 350  $\mu$  there was a change from a thickness of 115  $\mu$  to one of 205  $\mu$  and back again to 115  $\mu$ . From these anatomical studies it may be concluded that the mosaic disease affects the leaf tissues by causing a general increase in thickness in the dark green areas, and in the chlorotic areas a hypoplastic condition, accompanied by a decrease in differentiation of the palisade cells as well as a decrease in the number of chloroplasts and in their size.

Cytological studies were also made in order to determine whether or not there were present in the chlorotic cells any structures comparable to the bodies described in tobacco and petunia. A study of 100 sections of healthy leaves of *Datura* revealed no unusual inclusions, the cells all appearing perfectly normal. Likewise, there were no bodies found in 100 sections of the dark green areas of leaves of diseased plants. However, a study of the chlorotic areas revealed certain more or less irregular and indefinite structures, particularly in the upper epidermal cells.

These bodies resembled those described by Kunkel in corn much more nearly than they did those which are present in tobacco and petunia. They showed a strong affinity for Orange G, and at first sight gave the appearance of cytoplasm precipitated around the nucleus. Further study, however, demonstrated that they were a type of intracellular inclusion found only in the chlorotic areas of the mosaic-infected leaves. They were always adjacent to or surrounding the nucleus, showed

SMITH-MOSAIC DISEASES AND LEAF VARIEGATIONS

nothing of the nature of a limiting membrane, and were irregular in shape, size, and outline. Usually they were uniformly granular as in pl. 13, fig. 13, but occasionally they were more vacuolate as in pl. 13, fig. 15. In the vacuolate bodies, dark blue-staining granules could be observed as in fig. 15. Structures, such as the one illustrated in pl. 13, fig. 16, were apparently young stages in the development of the bodies, being small, taking the stain very lightly, and showing a tendency to be vacuolate. An unusual condition was found in which the nucleus was completely surrounded by the body, which last in turn was attached to the cytoplasm (see pl. 13, fig. 17).

At the time when the studies on the fixed material were made, no living diseased plants of *Datura* could be found, and consequently studies on living material have not yet been made. The author, however, feels that the bodies described here are definite entities which are associated with the chlorotic areas, since they were found only in those regions. When fresh material can be obtained observations will be made in order to see the appearance of these structures in the living cells.

4. Phytolacca decandra Linn.—Healthy and mosaic-infected poke were studied both in fixed material and in the living condition, the anatomical investigations being conducted as were those in the leaves of Datura. The following figures in each case are the average of 50 measurements which were made on the fixed material.

TABLE VIII

COMPARATIVE MEASUREMENTS OF TISSUES IN HEALTHY AND MOSAICINFECTED POKEWEED

	Healthy	Green area of mosaic	Chlorotic area of mosaic
Thickness of leaf	148 µ	191.5 μ	117.5 μ
Thickness of upper epidermis	15 μ	16. μ	15.5 μ
Thickness of palisade	57 μ	64.4 μ	27.3 μ
Thickness of spongy mesophyll	68.5 µ	100.2 µ	64.2 µ
Thickness of lower epidermis	7.5 µ	10.9 µ	11.0 4
No. of rows of palisade cells	1.	1.	1.
No. of rows of mesophyll cells	±4.	±4.	4.
Longest diameter of chloroplasts	5. μ	5. μ	5. μ

Here, as in Datura, it can be seen that the mosaic disease modifies the entire leaf, the dark green area being thicker than 5. Aquilegia.—Since 1919 Duggar has been observing a mosaic infection on plants of Aquilegia at the Missouri Botanical Garden. The diseased plants develop the typical mosaic symptoms including leaf mottling (see pl. 16, fig. 46), dwarfing of the plant, and decreased flower production. His inoculation experiments have indicated that it can be more or less readily transmitted. The infection was found on Aquilegia caerulea James, and since none of the healthy material of this species could be found, leaves of the hybrid A. canadensis Linn.  $\times$  A. californica Gray were used for comparison with the plants infected with mosaic. Anatomical studies were made as in Datura and in pokeweed, and the measurements tabulated in a similar manner, as follows:

TABLE IX

COMPARATIVE MEASUREMENTS OF TISSUES IN HEALTHY AND MOSAICINFECTED AQUILEGIA

	Healthy	Green Area of mosaic	Chlorotic area of mosaic
Thickness of leaf	110 μ	150 μ	100 μ
Thickness of upper epidermis	15 μ	20 μ	20 μ
Thickness of palisade	$42.5\mu$	50 μ	29 μ
Thickness of mesophyll	38.5 µ	57.5 μ	32.5 µ
Thickness of lower epidermis	15 μ	20 μ	20 μ
No. of rows of palisade cells	2	2	1
No. of rows of mesophyll cells	±4	±4	±4

These measurements, together with the semi-diagrammatic drawings in text-fig. 4, show that the most striking difference between the chlorotic and the dark green areas is a loss of one of the palisade layers in the former; although there is also a decrease in the thickness of the mesophyll, that in the dark green area being 1.76 times as great as that in the chlorotic sections. Here, again, the healthy leaves are thinner than the dark green areas and thicker than the chlorotic regions. The chloroplasts show a gradual degeneration in the transitional areas between the dark green and chlorotic areas, as shown in fig. 3 of text-fig 4. This loss of an entire palisade layer in the chlorotic areas is similar to the condition described by Funaoka ('24) in some of the variegations, e. g., that in Richardia Elliottiana, and Euphorbia marginata, in which there is a loss of certain layers of tissue in the chlorotic areas. The chlorotic appearance is therefore due

to the absence of one of the palisade layers as well as to a decrease both in the number of plastids and the chlorophyll content of these.

Cytological studies were also made on the living and the fixed material of both healthy and mosaic-infected leaves, but, although 100 sections were studied of each, there were no inclusions of any

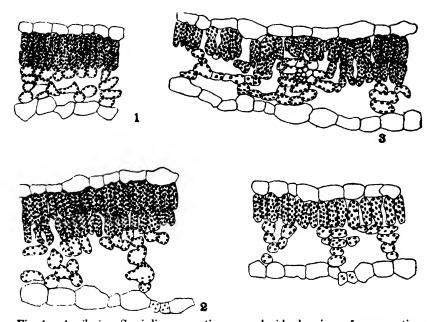


Fig. 4. Aquilegia. Semi-diagrammatic camera-lucida drawings of cross-sections of leaves, showing the effect of mottling on the tissue.  $\times$  300.

1. Aquilegia californica  $\times$  A. canadensis, healthy leaf; 2. A. caerulea, dark green area of an infected leaf; 3. A. caerulea, transitional area between chlorotic and dark

green areas of an infected leaf; 4, A. caerulea, chlorotic area of an infected leaf.

sort found in the chlorotic areas which did not also exist in the healthy and in the dark green portions.

From these studies on the tissues of healthy and mosaicinfected plants of Datura, pokeweed, and Aquilegia, it has been shown that the virus causes 2 changes in the leaves. apparently stimulates certain areas to a general increase in thickness, and second in other sections of the leaves it causes a marked inhibition of growth, the reduction being chiefly in the palisade cells, and in the number and size of plastids, and the amount of chlorophyll which they contain. The transitional

areas may follow the veins and be fairly sharp, as in *Datura*, or they may not be necessarily associated with the vascular system, at which times they are very gradual, as in pokeweed. Associated with the chlorotic areas in tobacco, petunia, and *Datura*, possibly also in pokeweed, there were certain irregular vacuolate or granular bodies which at times were associated with the nucleus, as in *Datura*. and at other times distributed in the cells independently of the nucleus. However, no such inclusions were found in the tissues of *Aquilegia* which were studied, showing that perhaps they do not universally accompany the mosaic infection in all plants.

#### **VARIEGATIONS**

It has been suggested that the infectious chloroses of Baur ('04, '06, '07, and '08) were of the same nature as the mosaic diseases, differing only in the difficulty with which they are transmitted. In this connection, therefore, it was of interest to make cytological studies of some of his variegations in order to determine whether inclusions comparable to those described above in certain of the mosaic infections could be found. was also a possibility that these bodies generally accompanied chlorotic tissues, arising perhaps from the disintegrating chloroplasts, hence were causally related to the chlorosis rather than directly induced by the action of the virus, or the causal agency. Therefore, studies were made on living and fixed material of various non-infectious variegations as well. A preliminary survey was made of many of those which were at the author's disposal, and from these the following were selected for more intensive study because they represent families in both the Monocotyledons and the Dicotyledons, because there are among them both infectious and non-infectious chloroses, and because they exhibit very different and distinct anatomical variations which, in some cases, are entirely different from those found in the mosaic plants infected with mosaic which have been studied.

1. Homalomena cordata Schott.—The variegation in this species consists of completely chlorotic round spots which, from microscopic observations, do not follow the veins, as shown in pl. 15, fig. 44. Free-hand sections were studied in the living condition, as well as sections of material which had been killed in chrom-

acetic acid, sectioned, and stained with Haidenhain's iron alum haematoxylin and Orange G.

Anatomical studies showed no difference in tissue differentiation between the green and the chlorotic areas. In neither region were the palisade cells developed, the leaf sections merely consisting of the upper epidermis, approximately 6 layers of spongy mesophyll, and the lower epidermis. The chief difference was the number and size of the chloroplasts; in the green areas, they were exceedingly large (averaging 8  $\mu$  in the longest diameter) and were more abundant, particularly in the upper layer of the spongy mesophyll which corresponds to the palisade layer in the ordinary leaf. On the contrary, the chloroplasts in the chlorotic areas were smaller and more sparsely distributed. The transitional region between the two areas was not sharp, the decrease in number and size of the plastids being very gradual. The nuclei in both the green and the chlorotic sections were strikingly large, both in the living and fixed materials.

Cytological studies, however, revealed no inclusions comparable in any way to those found in the plants infected with mosaic, although 100 sections of the fixed tissues and many living free-hand sections were examined. The living cells of both the green and chlorotic areas were frequently found filled with highly refractive bodies approximately 5  $\mu$  in diameter which were constantly in motion. Nevertheless, it is highly improbable that these bodies have any causal relation to the variegation, since they were found equally abundant in both the green and the chlorotic area. They were not observed in the fixed material.

2. Ficus Parcellii Veitch.—The variegation here is a mosaic-like mottling as shown in pl. 15, fig. 39. The different areas were very distinct, angular in appearance, and exhibited various distinct shades of green as well as pure white. Both living and fixed material were studied. Tissue taken from a nearly green leaf, such as that in pl. 15, fig. 45, was found to consist of the following layers of tissue,—upper epidermis, 2 layers of palisade cells, 4 layers of spongy mesophyll, and the lower epidermis, all of which contained chloroplasts. A study of the variegated leaves showed that the different colors of green had distinct

anatomical explanations, being due to a difference in distribution of the chloroplasts in the various layers. Transitional areas were always very sharp, one cell containing numerous chloroplasts, while the adjacent one might be completely devoid of them. It was found that there were 4 possible variants, as follows:

- a. First palisade layer.
- b. Second palisade layer.
- c. First two rows of spongy mesophyll cells.
- d. Lower two rows of spongy mesophyll cells.

Any one or any combination of these variants might be chlorophyll-free, while the remainder would be filled with chloroplasts. Some of the combinations which were observed are the following:

- 1. a, chlorophyll-free; b, c, and d filled with plastids.
- 2. b, chlorophyll-free; a, c, and d filled with plastids.
- 3. c, chlorophyll-free; a, b, and d filled with plastids.
- 4. b and d, chlorophyll-free; a and c filled with plastids.
- 5. d, chlorophyll-free; a, b, and c filled with plastids.
- 6. a, b, and d, chlorophyll-free; c filled with plastids.
- 7. a and b chlorophyll-free; c and d filled with plastids.

There was therefore no tissue differentiation between the green and the chlorotic areas, the difference in color being entirely due to an absence of plastids in one or more layers of tissue.

Cytological studies of both living and fixed cells revealed no inclusions comparable to those associated with the mosaic diseases.

3. Bougainvillea glabra Choisy.—The variegation in this species consists of a marginal chlorosis which may at times affect the entire leaf, as shown in the 3 leaves of pl. 16, fig. 48. The green portions of the variegated leaves, however, are not as dark a green as are the normal leaves, such as those shown in pl. 16, fig. 54. Microscopic studies showed that this had an easily interpreted anatomical explanation, which was similar to that given for the various shades of green found in Ficus Parcellii. The normal green leaf consists of an upper epidermis, one layer of palisade cells, 5 layers of mesophyll, and the lower epidermis, with chloroplasts distributed throughout the palisade as well as the spongy mesophyll cells. In the variegated leaves,

however, chloroplasts were never present in the palisade cells, being distributed only in the upper two layers of spongy mesophyll conthrough the entire mesophyll other than the palisade. The chlorotic areas lacked all chlorophyll. Here, again, the transition between the green and chlorotic areas was sharp, one cell containing normal plastids and the cell adjacent lacking them completely. Such transitions usually occurred near the veins. There was no difference in tissue differentiation between the two areas, the leaves being of uniform thickness.

Cytological studies on both living and fixed material gave no evidence of any intracellular inclusions which might be interpreted; as the causal agency of the chlorosis.

4. Pittosporum Tobira Ait.—The variegated leaves are a duller green than the normal leaves and have a completely chlorotic margin of varying depth, as shown in pl. 15, fig. 40, which may be compared with the green leaf in pl. 15, fig. 41. Here the normal green leaves consist of a thick upper epidermis, 2 or 3 layers of palisade tissue, 8 or 9 layers of spongy mesophyll, and a lower epidermis. The chloroplasts are distributed throughout the palisade and the spongy mesophyll, being more numerous in the former. In the green areas of the variegated leaves the distribution of the plastids is different, there being 2 layers of chlorophyll-free cells immediately below the upper epidermis and another 2 rows of similar cells immediately above the lower Accordingly, there may be said to be a chlorotic mantle surrounding the green tissue, thus accounting for the duller green color of the leaves. Funaoka ('24) has termed this type of chlorosis "periclinal variegation" and has described it in variegations of Pelargonium zonale, Glechoma hederacea, Acer Negundo, etc.

The variegated leaves show a white margin which is sharply set off from the dull green tissue. A microscopic study of this white area showed a notable decrease in thickness of the leaf, both the palisade layers and the spongy mesophyll being reduced. Particularly were the intracellular spaces reduced. In such areas the chlorophyll was completely lacking.

Cytological studies were made on both fixed and living material, and in neither the green nor the chlorotic areas could any unusual intracellular inclusions be observed in the 100 sections examined.

5. Nerium Oleander Linn.—Here, as in Pittosporum Tobira, the variegation consists of a chlorotic margin of varying depths, as shown in pl. 15, fig. 35, which is to be compared with the totally green leaves as shown in pl. 15, fig. 36. Microscopic studies show that the normal green leaf and the green portions of the variegated leaves are similar, except that in the entirely green leaf there are more chloroplasts in the lower part of the spongy mesophyll than there are in the variegated leaves. The green tissue consists in the order mentioned of an upper epidermis, 2 rows of chlorophyll-free cells, 2 rows of palisade cells, 6 rows of spongy mesophyll, 2 rows of chlorophyll-free cells, and a lower The chlorotic areas show the same tissue differentiaepidermis. tion, but the chloroplasts are replaced by leucoplasts throughout the spongy mesophyll and the palisade cells. The boundaries between the green and the chlorotic areas are very sharp, one cell showing the normal chloroplasts and the one adjacent exhibiting only leucoplasts. These transitions are always coincident with the veins. Occasionally, definite areas are observed which are lighter green than the normal, and this was found to be due to the fact that such areas lack chloroplasts in the upper palisade layer or in the spongy mesophyll cells.

Living and fixed tissues were studied cytologically but no unusual inclusions were found in either the green or the chlorotic areas.

6. Coleus Blumei Benth.—It was considered that perhaps the variety "Mrs. Kirkpatrick" of Coleus, fig. 37, pl. 15, was closely related to the mosaic infections, since it showed the pronounced crinkling of the leaves which is so characteristic of the mosaic symptoms. Sections of the leaves were studied in both the fixed and the living condition, but no intracellular inclusions other than degenerated chloroplasts could be found. In the fixed material the chloroplasts occasionally assumed appearances comparable to the vacuolate bodies in the tobacco and petunia mosaics, but they did not give the proper staining reactions, becoming more blue than orange. In the living cells they were not at all comparable to the vacuolate bodies in tobacco and would never be confused with them. Clear, highly refractive

bodies were seen in the living cells and were in constant motion as though exhibiting Brownian movement. They were found, however, in both the green and the chlorotic areas and could not be interpreted as a causal factor in the variegation.

Anatomical studies showed a condition different from any which have been described in this paper. The transition between the green and the chlorotic areas was so gradual as to be scarcely perceptible. The green area possessed two rows of palisade cells which gradually became shorter and thicker until they could no longer be distinguished from the spongy mesophyll. Therefore, in the chlorotic areas there was no differentiation between the palisade and the spongy mesophyll.

7. Evonymus japonica Linn.—There were two different variegations of this species at the author's disposal, both of which were studied by Baur. The variety "medio-picta" is shown in fig. 49 of pl. 16, as contrasted with the normal green in fig. 50 and the other variegation, "argenteo-variegata," in fig. 51. Anatomical studies revealed similar tissue differentiation in the 2 variegated varieties, so it will be necessary to include only a discussion of one of them, var. "medio-picta."

In this variegation the difference in tissue differentiation is found to be quite similar to that described for the *Datura* plants infected with mosaic except that here there are 3 layers of palisade cells. Measurements were made and tabulated as in the study of the plants infected with mosaic.

TABLE X

COMPARATIVE MEASUREMENTS IN GREEN AND VARIEGATED EVONYMUS LEAVES

	Normal green	Variegated green	Variegated chlorotic
Thickness of leaf	215 μ	420 µ	320 µ
Thickness of upper epidermis	20 μ	25 μ	20 μ
Thickness of palisade	150 μ	175 µ	100 μ
Thickness of spongy mesophyll Thickness of lower epidermis	$egin{array}{c} 125~\mu \ 20~\mu \end{array}$	200 μ 20 μ	185 μ 15 μ

These data show that the green leaves are much thinner than either the chlorotic or green portions of the variegated leaves, and that the difference lies chiefly in the palisade layer, which

is 3.5 times greater in the green areas of the wariegated leaves than in the green leaves. In the chlorotic areas the palisade layers are greatly reduced and contain relatively few chloroplasts, the ones that are present showing signs of disintegration. The transitional area between two regions usually covers about 5 or 6 cells.

Cytological studies were made on both the living and the fixed material of both types of variegations and of the mormal green plant. The chlorotic areas of both vars. "media-picta" and "argenteo-variegata" showed vacuolate bodies comparable to those found in tobacco and petunia mosaics, except that they occurred only in mesophyll and never in epidermal cells. Although at least 100 sections of the normal green tissue were studied, no such inclusions were found in these at any time.

The bodies were very similar in appearance to those in tobacco infected with mosaic. They showed a strong affinity for Orange G, contained several large or numerous small vacuoles, could be found adjacent to, or independent of, the nucleus, and occasionally, in the fixed material, exhibited numerous dark blue-staining granules (see pl. 14, figs. 25-32). No indication of a limiting membrane could at any time be observed. In the living cells the bodies appeared very similar to those in the fixed material, except that granules were never observed in any of the vacuoles.

Therefore, although these vacuolate bodies were not found in the non-infectious variegations studied, they were observed in the variegated varieties of *Evonymus japonica*. This is one of the species in which Baur ('08) found infectious chloroses. This would, then, appear to be evidence in favor of the view that these cell inclusions are associated directly with the virus rather than with the chlorosis which results from the presence of the virus, since they have been found in connection with only the one type of chlorosis—the infectious type. However, the author favors the view that they are not the causal agency itself but rather the product of a reaction between the virus and the cytoplasm of the cells.

## SUMMARY

- 1. Epidermal and hair cells of leaves of tobacco plants infected with the mosaic disease were examined in living and fixed tissues. The following observations of Goldstein were confirmed: the vacuolate bodies were not associated with the nuclei but were carried through the cells in the protoplasmic streams, the plate-like crystals were independent of the nuclei, and they lost their typical structure when placed in chrom-acetic acid. Contrary to her observations, however, the writer failed to observe any autonomous movements in the vacuolate bodies, and only once could the appearance of a limiting membrane be identified.
- 2. The observations of Rawlins and Johnson with reference to the fact that the inclusions occurred more frequently in greenhouse plants than in those grown out of doors were confirmed.
- 3. Treatment of mosaic-infected tobacco plants with the longer or biological ultra-violet rays for 18 days caused a dwarfing of the plants and a masking of the symptoms. Cytological studies of the rayed plants showed that the inclusions were present as in the controls, and these observations led to the conclusions that the absence of bodies in plants grown out of doors is not associated with the ultra-violet rays which they receive from the sun's spectrum.
- 4. The filtered mosaic tobacco juice was inactivated by an exposure of 30 minutes to the abiotic rays, whereas, under similar treatment, a suspension of *B. prodigiosus* was killed in 30 seconds. This is considered as evidence against the theory that the causal agency is an organism.
- 5. Living epidermal and hair cells of *Petunia* presented excellent material in which to study the intracellular inclusions. In cells showing rapid streaming, the vacuolate bodies exhibited 2 different movements: a migration through the cell, due to their being carried in the streams; and a change in form. The latter movement was explained as resulting from a combination of the effect of the force exerted on the mobile body by the streaming protoplasm and the apparent changes in form due to its turning over in the streams.
  - 6. Regarding the reactions to solvents, the vacuolate bodies

and the plate-like crystals were alike in their relatively high resistance to the action of formalin, HCl, and HNO<sub>2</sub>, and in the solubility in KOH. They differed, however, in the fact that the crystals were soluble in 10–95 per cent alcohol and 1–10 per cent acetic acid, whereas the vacuolate bodies were soluble only in 95 per cent alcohol and were not touched by acetic acid.

- 7. Anatomical studies of mosaic-infected leaves of *Datura*, pokeweed, and *Aquilegia*, showed that the virus enhanced the development of some areas and inhibited it in others, neither area, therefore, being normal. The reduction of tissue in the chlorotic regions was localized particularly in the palisade layers, and the chlorosis was accompanied by a decrease in size and number of chloroplasts.
- 8. Cytological studies of *Datura* revealed in the chlorotic areas irregular, granular, and vacuolate bodies in association with the nuclei, comparable to those described in corn by Kunkel. They were not found in the dark green areas of the diseased leaves or in the healthy tissues.
- 9. Cytological studies of pokeweed revealed intracellular vacuolate and sometimes granular bodies only occasionally present in the chlorotic areas.
- 10. No inclusions, not also present in the healthy cells, were found in the chlorotic areas of the diseased Aquilegia.
- 11. Anatomical studies were made on seven different variegations, and the variations in the difference between the chlorotic and green areas in the various types of chloroses were observed.
- 12. No inclusions were found associated with the chlorotic areas in *Homalomena cordata* Schott, *Ficus Parcellii* Veitch., *Bougainvillea glabra* Choisy var. variegata, *Pittosporum Tobira*, Ait. var. "variegatum," Nerium Oleander Linn., and Coleus Blumei Benth. var. "Mrs. Kirkpatrick."
- 13. Cytological studies revealed the presence of vacuolate bodies in the mesophyll cells of the chlorotic areas of *Evonymus japonica* vars. "argenteo-variegata" and "medio-picta." Evonymus japonica is one of the species in which Baur found infectious chloroses.
- 14. The vacuolate bodies, therefore, have been observed, as yet, only associated with the infectious chloroses.

15. These observations seem to justify the conclusion that the vacuolate and granular bodies discussed in this paper are associated directly with the causal agency rather than with the chlorosis which results from the presence of the virus in the plant. The author favors the view, however, that they do not represent the causal agency, but are rather the product of a reaction between it and the cytoplasm of the cells.

### ACKNOWLEDGMENTS

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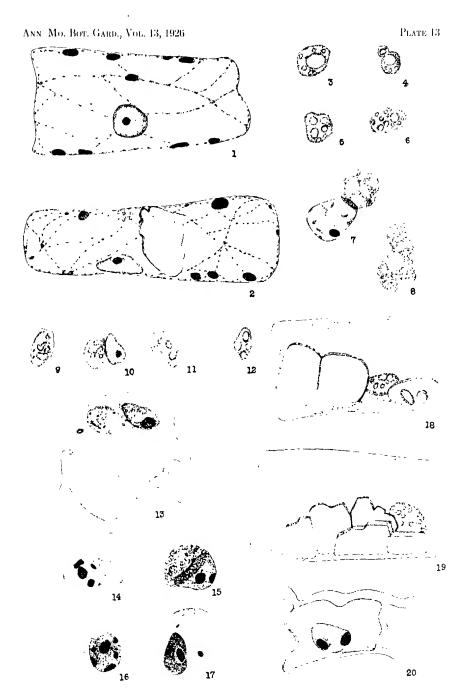
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#### PLATE 13

- Fig. 1. Petunia. Living hair cell from leaf of healthy plant, showing only a nucleus and small plastids, and no inclusions. X 1000.
- Fig. 2. Petunia. Living hair cell from the chlorotic area of a mosaic-infected leaf, showing, in addition to the nucleus and plastids, a typical vacuolate body near the plate-like crystal. × 1000.
- Figs. 3-6. Petunia. Vacuolate bodies as seen in the living hair cells and epidermal cells. × 1000.
- Fig. 7. Petunia. Rounded vacuolate body adjacent to the nucleus in an upper epidermal cell of the chlorotic area of a mosaic-infected leaf. The tissue was fixed in chrom-acetic acid, and stained with Haidenhain's haematoxylin and counterstained with Orange G. No formed structures or limiting membrane were present. × 2000.

# EXPLANATION OF PLATE (Continued)

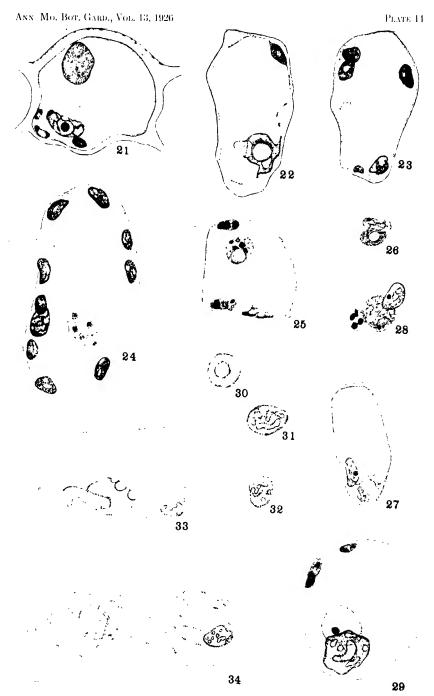
- Fig. 8. Petunia. Irregular amoeboid-like vacuolate body in a mesophyll cell immediately above the lower epidermis. Fixed and stained as in fig. 7. × 2000.
- Fig. 9. Petunia. Vacuolate body as seen in a hair cell which had been kept in 15 per cent alcohol for a 12-hour period. The resulting shrinkage left visible a distinct limiting membrane. × 1000.
- Fig. 10. Petunia. Nucleus and adjacent body of a cell which had been held in 30 per cent alcohol for 12 hours, showing that this concentration of alcohol neither dissolved nor materially modified these vacuolate bodies. × 1000.
- Fig. 11. Petunia. Vacuolate body in a hair cell which had been kept in 50 per cent alcohol for 12 hours, showing that there were no injurious effects. × 1000.
- Fig. 12. Petunia. Vacuolate body in a hair cell which had been placed in 70 per cent alcohol for 12 hours, showing no solution nor disintegration even at this high concentration. × 1000.
- Fig. 13. Datura Stramonium. Upper epidermal cell in chlorotic area of a mosaic-infected leaf, showing the indefinite granular body adjacent to the nucleus. Several dark-staining granules are present in it. The leaf was fixed in chrom-acetic acid and stained in Haidenhain's iron alum haematoxylin and counterstained with Orange G. × 2000.
- Fig. 14. Datura Stramonium. Nucleus and adjacent body in an upper epidermal cell in the transitional area between the chlorotic and green area in fixed material of a mosaic leaf. The body shows some suggestion of a vacuolate structure, but no limiting membrane. × 2000.
- Fig. 15. Datura Stramonium. Nucleus with adjacent body in an upper epidermal cell of the chlorotic area in a mosaic-infected leaf. The material was fixed and stained as in fig. 13. This is a rather typical appearance of the body, being filled with small vacuoles in which there were occasionally dark-staining granules.  $\times$  2000.
- Fig. 16. Datura Stramonium. Apparently the young stage of a body. It was found in the upper epidermal cell in the chlorotic area of a mosaic-infected leaf which had been fixed and stained as in fig. 13.  $\times$  2000.
- Fig. 17. Datura Stramonium. Unusual appearance of the body which completely surrounded the nucleus and was attached to the cytoplasm at the edge of the cell. Upper epidermal cell of chlorotic area. The body is less dense than usual and contains several dark-staining granules.  $\times$  2000.
- Fig. 18. Petunia. Portion of hair cell from the chlorotic area of a mosaic-infected leaf, which had been placed in 4 per cent formaldehyde for a 12-hour period. The vacuolate body was still normal in appearance. The crystals had lost their sharp angular edges, and in the one on the right of the body disintegration and solution had set in. × 1000.
- Fig. 19. Petunia. Portion of a hair cell from the chlorotic area of a mosaic-infected leaf which had been kept in 4 per cent formaldehyde for 1 hour. Both the vacuolate body and the plate-like crystals were normal in appearance. × 1000.
- Fig. 20. Datura Stramonium. Healthy epidermal cell fixed in chrom-acetic acid and stained with Haidenhain's iron alum haematoxylin and Orange G. No inclusions were present. × 2000.



SMITH MOSAIC DISLASES AND LEAF VARIEGATIONS

#### PLATE 14

- Fig. 21. Phytolacca decandra. Upper epidermal cell of the chlorotic area of a mosaic-infected leaf, showing the very regular rounded body containing numerous small vacuoles, some of which surround very small dark-staining granules. The leaf was fixed in chrom-acetic acid and stained with Haidenhain's iron alum haematoxylin and Orange G. × 1500.
- Fig 22. Phytolacca decandra. Cell immediately above the lower epidermis in the chlorotic area of a mosaic-infected leaf, fixed and stained as in fig. 21. This most unusual body was vacuolate, took a light gray-blue stain, was only slightly denser than the cytoplasm, and apparently was not associated with the nucleus. × 1500.
- Fig. 23. Phytolacca decandra. Cell just above the lower epidermis in the chlorotic area of a mosaic-infected leaf, fixed and stained as in fix. 21. The cell showed the typical appearance of the bodies, the latter being a clear blue-gray and containing several large vacuoles. Several chloroplasts were also present in the cell. × 1500.
- Fig. 24. Phytolacca decandra. Cell in second row above the lower epidermis in the chlorotic area of a mosaic-infected leaf. The very definitely walled structure in the center of the cell took a deep orange stain in the periphery which surrounded and gradually changed into the dark blue-staining centers. In addition, the cell contained 9 chloroplasts and a nucleus. × 1500.
- Fig. 25. Evonymus japonica. var. "medio-picta." Mesophyll cell in the chlorotic area of the variegated leaf. The body contained a large vacuole, numerous small ones, and several dark-staining granules. It apparently was not associated with the nucleus which was in the lower part of the cell among several plastids. × 1500.
- Fig. 26. Evonymus japonica var. "medio-picta." Nucleus and adjacent body which were found in a cell next to the one in fig. 25. The body contained a single large vacuole with several small ones. The material was fixed in chrom-acetic acid and stained with Haidenhain's iron alum haematoxylin and Orange G. × 1500.
- Fig. 27. Evonymus japonica var. "argenteo-variegata." Mesophyll cell in the chlorotic area of the variegated leaf, containing only the nucleus and a vacuolate body. There were no dark-staining granules in the body, and only 2 large vacuoles but many small ones. The material had been fixed and stained by the usual method. × 1500.
- Fig. 28. Evonymus japonica var. "argenteo-variegata." Vacuolate body in a mesophyll cell of the chlorotic area of a variegated leaf. It was surrounded by the nucleus and 2 disintegrating plastids. It contained two large vacuoles with dark-staining granules, and numerous small ones. The material was fixed and stained as in the previous figures. × 1500.
- Fig. 29. Evonymus japonica var. "medio-picta." Living mesophyll cell showing the nucleus with the vacuolate body partially superimposed. Two small chloroplasts were present in the cytoplasm.  $\times$  1500.
- Figs. 30, 31, and 32. Evonymus japonica var. "medio-picta." Vacualate bodies as seen in the living mesophyll cells in the chlorotic areas of the variegated leaves  $\times$  1500.
- Fig. 33. Petunia. Portion of a hair cell from the chlorotic area of a mosaic-infected leaf which has been placed in 8 per cent formalin for 1 hour, showing the normal appearance of the vacuolate body but the beginning of disintegration of the crystals. × 750.
- Fig. 34. Petunia. Hair cell from the chlorotic area of a mosaic-infected leaf which had been kept in 10 per cent HCl for 6 hours, showing that, although there had been very severe plasmolysis, neither the crystals nor the body were injured. × 750.



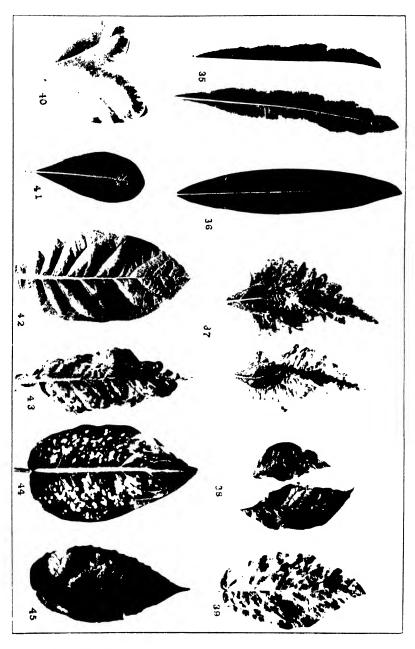
SMITH MOSAIC DISPASES AND LEAF VARILGATIONS

# PLATE 15

### (Leaves one-half natural size)

	· · · · · · · · · · · · · · · · · · ·
Fig. 35.	Leaves of the variegated variety of Nerium Oleander Linn.
Fig. 36.	Normal green leaf of Nerium Oleander Linn.
Fig. 37.	Leaves of Coleus Blumei Benth. var. "Mrs. Kirkpatrick."
Fig. 38.	Mosaic-infected poke leaves.
Fig. 39.	Variegated leaf of Ficus Parcellii Veitch.
Fig. 40.	Leaves of Pittosporum Tobira Ait. var. variegatum.
Fig. 41.	Normal green leaf of Pittosporum Tobira Ait.
771 40	77 1.1

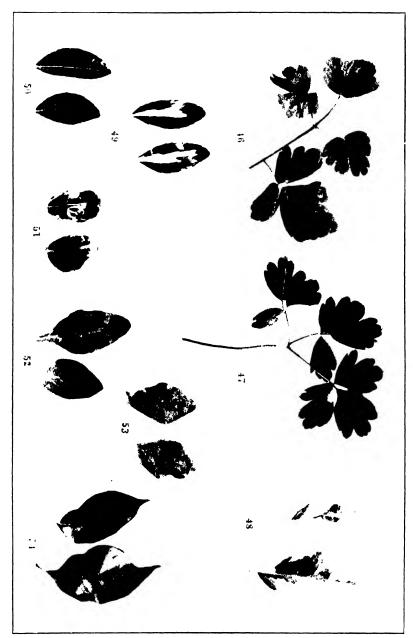
- Fig. 42. Healthy tobacco leaf.
- Fig. 43. Mosaic-infected tobacco leaf.
- Fig. 44. Leaf of Homalomena cordata.
- Fig. 45. Nearly entirely green leaf of Ficus Parcellii Veitch.



### PLATE 16

### (Leaves two-thirds natural size)

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- Fig. 47 Leaves of healthy Aquilegia.
- Fig. 48. Leaves of Bougainvillea glabra Choisy var. variegata.
- Fig. 49. Leaves of Evonymus japonica var. "medio-picta."
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